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Initial Characterisation of Lakes Prespa, Ohrid and Shkodra/Skadar

Implementing the EU Water Framework Directive in South-Eastern Europe

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Rruga Skenderbej
Pallati 6, Ap. 1/3
Tirana, Albania
T +355 42 258 650
F +355 42 251 792
www.giz.de

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Text
Emirjeta Adhami, Ariola Bacu, Sajmir Beqiraj, Pavle Djurašković, Aleksandar Flloko, Dafina Gusheska, Sead Hadžiablahović, Dushica Ilik-Boeva, Ekerem Kapedani, Lefter Kashta, Goce Kostoski, Xhume Kumanova, Lence Lokoska, Danilo Mrdak, Dragana Milošević, Arian Palluqi, Arben Pambuku, Suzana Patceva, Ana Pavićević, Marash Rakaj, Jelena Rakočević, Elizabeta Veljanoska Sarafiloska, Zoran Spirkovski, Spase Shumka, Marina Talevska, Trajche Talevski, Orhideja Tasevska, Sonja Trajanovska, Sasho Trajanovski

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Contents

Introduction

The present Volume of Annexes contains original reports of water quality investigations undertaken by partner institutions and experts with support of GIZ. The Volume complements the Initial Characterization Report (ICR) which in turn focuses on the main findings of the original work reported here or published elsewhere. Contrary to the ICR main report and in order to preserve their authenticity, the original reports have not been submitted to a rigorous review. Therefore, only gross errors or inconsistences were reported back to authors along with a request to revise their reports accordingly. However, content and text of the final reports were entirely under the responsibility of the authors while the Publisher was in charge of harmonizing layout and formatting style.

REPORT

on

Joint integrated monitoring program for the transboundary Lakes of Shkodra/Skadar, Ohrid and Prespa

Prepared by: Arben Pambuku, Ph.D.
Xhume Kumanova, Ph.D.
Emirjeta Adhami, Ph.D.

June 2014

Contents

1. Introduction

This report is written upon the request of the project "Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar (CSBL)". The report is focused on the monitoring of physic-chemical elements which has been agreed by the Technical Working Group WFD (TWG WFD) within the CSBL project. Furthermore sampling campaigns, analysis methods and the evaluation has been harmonized on transboundary levels to fulfill the WFD requirements.

When a new programme is being started, or a lapsed programme is being reinstated, it is useful to begin with a small-scale pilot project. This provides an opportunity for newly trained staff to gain hands-on experience and to confirm whether components of the programme can be implemented as planned. It may also provide an opportunity to assess the sampling network and provide indications of whether more (or possibly fewer) samples are needed in order to gain knowledge of the water quality at various points throughout a water body.

Within a pilot project like the CSBL approach for the three lakes it is important to test assumptions about the mixing of lakes at the selected sampling sites and times. Therefore it might be appropriate to consider variations in water quality at selected sampling sites throughout an annual cycle in order to confirm the number of samples required to produce representative data.

In a lake it may be necessary to sample at different points to determine whether water quality can be estimated at a single point or whether the lake behaves as a number of separate water bodies with different water quality characteristics. It is also essential to investigate variation in water quality with depth and especially during stratification. Lakes are generally well-mixed at overturn (i.e. when stratification breaks down) and sampling from a single depth or the preparation of a composite sample from two depths or more may adequately represent the overall water quality.

All these elements are taken into account when the investigation program of CSBL has been developed by the TWG WFD and harmonized by the experts of AGS and partners in Macedonia and Montenegro.

At this report is made the assessment of the results taken during the sampling campaign and analyzing in chemical laboratory by Mr. Arben Pambuku, Mrs. Xhume Kumanova and Mrs. Emirjeta Adhami.

Sampling campaigns have been carried out by AGS sampling team (Arben Pambuku, Joni Topulli, Megli Bele, Marenglen Gjoka). Analyses of samples have been done by AGS Laboratory staff (Xhume Kumanova, Suzana Qilimi).

2. Selection of the sampling points

Processes affecting water quality and their influence should be taken into account when sampling sites are selected. A sampling site is the general area of a water body from which samples are to be taken and is sometimes called a "macro-location". The exact place at which the sample is taken is commonly referred to as a sampling station or, sometimes, a "micro-location". Selection of sampling sites requires consideration of the monitoring objectives and some knowledge of the geography of the water-course system, as well as of the uses of the water and of any discharges of wastes into it. Sampling sites are marked on a map after a final decision on the precise location of a sampling station that is made by National Agency of Environment (from the STEMA project). The selected numbers of monitoring stations are selected to assess the magnitude and impact of the significant point source, diffuse source and hydro-morphological pressures.

Based on the identified significant pressures and the natural conditions AGS agreed the location of the sampling sites with TWG WFD.

2.1 Lake Shkodra

Lake Shkodra is the biggest lake of Balkan Peninsula. The water mirror surface of Shkodra Lake is 368 km^2; from which 149 km^2 are placed inside of Republic of Albania territory, while the other part is placed on Montenegro. It is a field lake, with low depths, which approximately varies from 7 to 10 m. The maximum depth of Shkodra Lake is 44 m. This lake presents such a kind of crypto depression. The Shkodra Lake waters discharge into Adriatic Sea via Buna river bed, which is and one natural channel of this lake. The total volume of Shkodra Lake water for the average multiannual level of this lake is 2.6 milliard m^3.

From geographical point of view, Shkodra Lake include at Adriatic watershed, where it discharges its waters of its hydrographic network via Buna River bed. Watershed of Shkodra Lake, border on north with watershed of Trebeshnica, Neretva, Piva and Tara rivers. Trebeshnica and Neretva River flow in Adriatic Sea, while Piva and Tara are brunch of Drina River first, after Sava River and at the end Danubi River, which flow in Black sea. At the SE direction the watershed of Shkodra lake border on Drini watershed and on the south direction it border with Albanian West Lowland, in which is spreading the bed of Buna River. On the west direction, it border with water of Adriatic Sea. Shkodra Lake gather waters from an area with general watershed surface around 5179 km^2, from which 1025.2 km^2 are included inside of Albanian territory, while the other part of it is at Montenegro area.

Following monitoring stations have been chosen for the monitoring programme in the Albanian part of the lake:

- Kalldrun (Sterbeq) station – at coordinates N 42° 11' 44" and E 19° 23' 0.29".
  It is in the northern part of the Albanian part of lake. The territory in this area has a considerable agricultural surface which is very adapted for plants such vegetables, tobacco, fodder, viticulture, ether plants, etc. Also, in this side of lake, fishery and livestock breeding are developed as an economic activity. These activities are in a non problematic level in the area.
- Zogaj station – it is in the southwest part of Albanian part of lake. In Zogaj area the main activities are agriculture and fishery. The agriculture is focus in olive yards, apiculture and cattle. The tourism is developed during last five years where some restaurants along the shore were built, only 2 hotels and 2-3 houses offer accommodation service.
- Shiroka station – at coordinates N 42° 04' 22.6" and E 19° 24' 13.9".
  It is in the southern part of Albanian part of lake. There is an urban pressure. At Shiroka area the main activity is fishery and small tourism mainly as small restaurant. Main impact on this part comes from urban discharges.

2.2 Lake Ohrid

Lake Ohrid is the deepest tectonic lake in the Balkans (289 m, 7th deepest lake in Europe). It covers in total an area of 358.2 km2 and only 109 km^2 belongs to Albania. The altitude of the Lake is 695 m a.s.l.

The coastline of Lake Ohrid is 87.5 km long, of which 56 km belongs to Macedonia and 31.5 km to Albania. The lake itself has a maximum length of about 30km and a width that varies from 11.2 km to 14.5km. The maximum depth is 289 m and average depth is 164 m.

The water level of Lake Ohrid has been gradually decreasing over time. The evidence of this is apparent in the terraces along the lakeshore. There are 2-3 distinct and perhaps as many as 5 terraces on some shorelines reflecting previous lake levels. On the north part of the lake in Macedonia there is a lake terrace under the surface of the lake. The terraces along the sides of the lake represent some of the most productive fields for agriculture (Bucimas, Lin, Struga, and Ohrid). The ancient isles of Lini and Ohri have turned into peninsulas. The field of Lini has clearly retained traces of its lake origin (wet grounds and the black colour of the alluvium).

Lake Ohrid is the deepest lake in Albania and in collaboration with the MoEFWA and NEA that offered environmental monitoring database it is decided to have three monitoring stations. The selection of station is based on the main pollution resource in the lake and also on the main pressures coming into lake from point and diffuse sources of pollution.

Following monitoring stations have been chosen for the monitoring programme in the Albanian part of the lake:

- Lin station – at coordinates N 41° 4' 7.95" and E 20° 38' 35.63". It is in the northern part of the

Albanian part of lake. The main impact is urban pressure and also from the fisheries.

➢ Memelishti station – at coordinates N 40° 55' 56.03" and E 20° 39' 1.26". It is under mineral damp pressure and urban pollution. To the northwest of Pogradeci, there are a number of old mines that used to produce chromium, nickel, iron, and coal. Only one of these remains in operation, but at the mining sites, many large piles of waste material remain and are a source of pollutants to the lake each time it rains.

➢ Tushemishti station – at coordinates N 40° 54' 18.39" and E 20° 39' 26.71". It is in the southern part of Albanian part of lake. There is urban and agriculture pressures. Also there are some tourism resorts that impact the shoreline. The agriculture in the area is characterized by a low intensity with limited use of pesticides and mechanic equipment, and includes crop production and livestock breeding. The lands used for agriculture are irrigated using Pogradeci Lake water via pumping stations. Agriculture is concentrated mainly in the territories of Hudenisht, Starove, and Çerrave Communes. About 1500 ha of cultivated land are irrigated using water from both the Drilon River and Lake Ohrid. Most of the drained water discharged directly or indirectly into the lake.

2.3 Lake Prespa

Prespa Lakes are a high-altitude basin (850 masl) with two inter-connected lakes: Micro Prespa (47.35 km2, shared between Albania and Greece) and Macro Prespa (259.4 km2, shared between all countries).

The overall basin catchment is quoted as 2,519.1 km2 although the complex karstic nature of the catchment has resulted in a range of values. The lake is surrounded by high altitude mountains reaching 2,601 m (Mount Pelister). Micro and Macro Prespa are connected by a short sluice-controlled water course which regulates the level of Micro Prespa. There are four main rivers feeding Macro Prespa with Micro Prespa receiving water from diverted Devoli River. In addition there are a number of ephemeral water courses. The outflow from Macro Prespa is believed to be to Lake Ohrid through the karstic geology. The selection of station is based on the main pollution resource in the lake.

Following monitoring stations have been chosen for the monitoring programme in the Albanian part of the lake:

➢ Gollomboc station – at coordinates N 40° 53' 0.08"and E 20° 55' 59.23". The pressure at this area comes from the agriculture activities and urban discharges.

➢ Pusteci (Liqenas) station – there is urban and agriculture pressure. The coast of lake at this area is flat and it is under the erosion pressure.

3. Description of the sampling campaigns

Water samples were taken at all sampling stations in four sampling campaigns which were belonged to four different seasons, as it is recommended of WFD and joint monitoring protocol: July, October 2013 and February, April 2014. Also a join sampling procedure applied at the pelagic station in Ohrid Lake with participants from Albania and Macedonia. The samples taken in this station were analysed in both laboratories (AGS and HBIO). The data about the sampling campaign put in monitoring field sheet where data from the location (coordination), weather conditions, water of lake visibility (transparency) and the parameters measured in the field were prescribed. Also samples from the Shkodra Lake are changed between Albanian and Montenegrin partners. They are analysed in both laboratories (AGS and IHMS).

According to the field sheet data the weather conditions during the field campaign were:

- ☞ *On July 2013* - good sunny weather and no wind, no wave
- ☞ *On October 2013* – weather was not good with wind and cloudy/rain, small waves
- ☞ *On February 2014* – snow, rain and low air temperature, high waves and windy
- ☞ *On April 2014* - good sunny weather and no wind, no wave

During the third sampling campaign the AGS sampling team has completed their work in difficult conditions accompanied with wind and high waves, low temperatures and snow/rain.

The field monitoring sheet are attached this report in Annex 1.

4. Methods for sampling, transport and storage, analysis

4.1 Sampling techniques

Collecting of samples was done by two motor-boats. One day was spent for collection of samples in each campaign for Shkodra and Prespa Lake and two days for the Ohrid lake samples for the Albanian team.

Samples of water were taken by a Ruthner bottle of 3 litres. Water samples were taken at two depths (surface and bottom of water column) in Shkodra Lake and at one depth (surface of the water column) in Ohrid and Prespa Lakes. All sampling procedures conducted according official standard methodology using field equipment. Samples for general chemical analysis were collected in containers of polyethylene bottles or glass bottle 1 litre. Samples were collected in Winkler bottles of 100 ml for dissolved oxygen and 250 ml for $BOD_{5.}$ Samples for chlorophyll-a were collected in dark glass bottles of 1.5 litres. Also samples for microelements (heavy and trace metals) collected in polyethylene bottles 1 litre and were treated with HNO_3 concentrated solution (1:1). All samples were registered putting the code of the sampling site, parameters that will be analysed and the amount of the sample according to the procedure of the registration and also the field sheet data were filled in site during the sampling campaign.

4.2 Preservation/treatment of the samples

According to the standard for preservation and treatment of samples AGS has used the table below for the physico-chemical parameters of water quality:

Parameter	Containers	Treatment and conservation	Minimal amount of sample (ml)	Maximal time for conservation
Alkalinity	Polyethylene	Cooling on 4°C	100	24 hour
BOD	Polyethylene	Cooling on 4°C	250	4 hour
TOC	Polyethylene	Cooling on 4°C	200	24 hour
Chloride	Polyethylene	Cooling on 4°C	100	7 day
Chlorophyll	Glass test tube with stopper (after filtration)	Freezing on -20°C	1500	7 day
COD	Polyethylene	Cooling on 4°C	200	24 hour
DO (Winkler)	Glass	Fixed in site	300	6 hour
NH_3	Polyethylene	Cooling on 4°C 2 ml H_2SO_4 40%/l	400	24 hour
NO_2+NO_3	Polyethylene	Cooling on 4°C	200+50	24 hour
pH	Polyethylene	Nothing	100	6 hour
Dissolved phosphate	Glass	Filtrate with filter 0.45mm	100	24 hour
Inorganic phosphorous	Glass	Cooling on 4°C	100	24 hour
Total phosphorous	Glass	Cooling on 4°C	100	1 month
Conductivity	Polyethylene	Cooling on 4°C	100	24 hour

Bottles with samples were disposed in cooling boxes, at low temperature. The samples were transported to laboratory by car. The containers with samples were saved from any outside influences. The samples carefully transported to laboratory as soon as possible from the sampling day. When the samples came in laboratory, they disposed in refrigerators at 4°C, waiting to analysis.

4.3 Methods of parameters/indicators measures

All analyses for water quality parameters were done according to the agreed standard methods. Temperature of water, depth, transparency by Secchi disk, pH, DO and conductivity were measured in the field by thermometer, multi-parametric analytic equipment that was calibrated before in the laboratory. The samples of chlorophyll **a** was filtered through the glass filter 0, 45 µm, as soon as they come in laboratory. The samples for dissolved oxygen were conserved with $MnCl_2$ and KJ, at the field and analyse of BOD_5 was complete after five days with the same procedure as for DO. All the other parameters analyzed in laboratory within determined time according to the specification methods of each parameter.

The parameters of water quality are measured in laboratory that use the standard methods like ISO, EN standards or methods taken from the American manual for water and waste water examination. The methods used by laboratory are presented in the table below:

Table 1. Table of method used by laboratories that will be part in monitoring program implementation for Shkodra, Ohrid and Prespa Lakes

Variable	Method	Equipment
Temperature	(APHA-AWA-WPCF, 2010)	Reversing thermometer
Transparency	(APHA-AWA-WPCF, 2010)	Secchi disc
pH	ISO 10523:2012	pH-meter WTW pH 197 Glass electrode. No stirring.
Conductivity	ISO 27888:1993	Conductometer
Alkalinity as phenolphthalein	S-SH 2639-22:1990	Titrimetry
Dissolved oxygen	(APHA-AWA-WPCF, 2010)	Titrimetry
COD-Mn	S-SH 2639-15	Titrimetry
BOD_5	(APHA-AWA-WPCF, 2010)	Titrimetry
Chloride (parameter for salinity)	ISO-10304-1:2007	Ion Chromatography
Sulphate	ISO-10304-1:2007	Ion Chromatography
NH_4-N	ISO-14911:2003	Ion Chromatography
NO_2-N	ISO-10304-1:2007	Ion Chromatography
NO_3-N	ISO-10304-1:2007	Ion Chromatography
Phosphate	ISO-10304-1:2007	Ion Chromatography
Total phosphorous	ISO-10304-1:2007	Ion Chromatography
TOC	S SH EN 1484:2000	Analyzer for TC and TN
Pesticides	(APHA-AWA-WPCF, 1998)	Gas Chromatography
Heavy metals (Cd, Pb)	AAS-Flame	Atomic Absorber

Interpretation

Results Lake Shkodra

Station AL 1 – Kalldrun (Bajza)

Parameters	Frequency	Unit of measurement	Surface	Depth	Surface	Depth	Surface	Depth	Surface	Depth
date/time of sampling			18/7/2013	18/7/2013	9/10/2013	9/10/2013	10/2/2014	10/2/2014	12/4/2014	12/4/2014
temperature	every 3 months	C	28.1	16.2	20.4	14.1	9.47	10.1	18.4	17.5
transparency	every 3 months	m	4	4	2.5	2.5	1.5	1.5	2.4	2.4
conductivity	every 3 months	µS/cm	221	253	225	226	267	282	267	267
pH	every 3 months		8.18	7.96	8.11	8.05	8.23	8.29	8.22	8.14
alkalinity as phenolphthalein	every 3 months	ml HCl	118	133	113	115	140	140	132.5	132.5
Total Suspended Solids - TSS	every 3 months	mg/l	3.3	12.2	<0.1	<0.1	<0.1	<0.1	0.6	32.43
Dissolved Oxygen - DO	every 3 months	mg/l	7.64	7.54	7.69	7.57	9.7	8.5	5.9	4.4
Oxygen Saturation - OS	every 3 months	% O_2								
COD	every 3 months	mg/l	0.8	0.8	0.84	0.84	0.76	0.6	0.56	0.10
BOD_5	every 3 months	mg O_2/l	2.44	2.63	2.52	2.75	1.73	1.95	1.7	0.6
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.02	0.02	0.03	0.06	0.02	0.04
NO_3-N	every 3 months	mg N/l	0.18	0.18	0.09	0.06	0.367	0.37	0.97	0.93
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.02	0.02
total nitrogen	every 3 months	mg N/l	0.33		0.21	0.24	0.343	0.461	0.29	0.32
ortho-phosphorus	every 3 months	µg P/l	30	20	30	20	29	26	16	20
TC	every 3 months	mg/l	1.85		3.62	4.46	3.58	4.54	3.35	4.18
Chloride	every 3 months	mg/l	3.55	8.88	5.33	3.55	2.272	2.272	2.16	3.32
Sulphate	every 3 months	mg/l	7	6.58	7.65	7.23	2.494	2.892	2.71	2.75
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	0.33				1.2			
Lead	one per year	mg/kg	<0.01	<0.01	<0.0		0.01			0.01
Cadmium	one per year	mg/kg	<0.001.	<0.001	0.001		0.001			0.001

Station AL 2 – Zogaj

Parameters	Frequency	Unit of measurement	Surface	Depth	Surface	Depth	Surface	Depth	Surface	Depth
date/time of sampling			18/7/2013	18/7/2013	9/10/2013	9/10/2013	10/2/2014	10/2/2014	12/4/2014	12/4/2014
depth	every 3 months	m	6	6	6	6	7	7	6	6
sampling depth	every 3 months	m	0	6	0	4	0	7	0	6
temperature	every 3 months	C	27.8	17	20	17.7	9.8	9.75	18.3	17.2
transparency	every 3 months	m	3		2.2		3		2.5	
conductivity	every 3 months	µS/cm	226	226	222	223	266	268	269	269
pH	every 3 months		8.36	8.28	8.45	8.46	8.3	8.26	8.28	8.29
alkalinity as phenolphthalein	every 3 months	ml HCl	125	123	113	113	138	135	132.5	135
Total Suspended Solids - TSS	every 3 months	mg/l	4.4	15.4	1.8	1.8	<0.01	<0.01	1.74	3.86
Dissolved Oxygen - DO	every 3 months	mg/l	7.92	7.42	7.7	7.52	10.2	9.8	11.2	10.4
Oxygen Saturation - OS	every 3 months	% O_2								
COD	every 3 months	mg/l	0.52	0.52	0.68	0.52	0.56	0.6	0.6	0.96
BOD_5	every 3 months	mg O_2/l	2.24	2.67	2.5	2.62	2.1	2.3	1.3	0.95
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.02	0.02	0.05	0.08	0.04	0.03
NO_3-N	every 3 months	mg N/l	0.54	0.18	0.2	0.07	0.295	0.286	0.21	0.2
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	0.01	<0.01	<0.01	<0.01	0.02	0.02
total nitrogen	every 3 months	mg N/l	0.22	0.19	0.2	0.18	0.301	0.362	0.26	0.33
ortho-phosphorus	every 3 months	µg P/l	10	10	10	10	31	15	20	10
TC	every 3 months	mg/l	2.19	2.17	3.67	3.45	3.76	4.48	3.88	4.75
Chloride	every 3 months	mg/l	3.55	3.55	5.33	5.33	2.227	2.695	2.13	2.36
Sulphate	every 3 months	mg/l	4.53	4.53	2.88	2.47	2.425	2.459	2.69	2.73
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	0.22				1.4			
Lead	one per year	mg/kg	<0.01	<0.01	<0.01		0.03			0.01
Cadmium	one per year	mg/kg	0.004	0.003	0.001		0.002			0.001

Station AL 3 – Shiroka

Parameters	Frequency	Unit of measurement	Surface	Depth	Surface	Depth	Surface	Depth	Surface	Depth
date/time of sampling			18/7/2013	18/7/2013	9/10/2013	9/10/2013	10/2/2014	10/2/2014	12/4/2014	12/4/2014
depth	every 3 months	m	4	4	4	4	4	4	4	4
sampling depth	every 3 months	m	0	4	0	4	0	4	0	4
temperature	every 3 months	C	28.3	27.5	21	16.5	9.33	9.2	17.9	17.4
transparency	every 3 months	m	3.5		2.7		3.5		2.4	
conductivity	every 3 months	µS/cm	224	226	226	226	262	271	269	267
pH	every 3 months		8.38	7.26	8.45	8.28	8.29	6.92	8.25	8.31
alkalinity as phenolphthalein	every 3 months	ml HCl	125	123	114	115	140	140	135	135
Total Suspended Solids - TSS	every 3 months	mg/l	3.4	1	3.7	3.1	0.8	<0.10	0.6	1.4
Dissolved Oxygen - DO	every 3 months	mg/l	7.79	7.22	7.81	7.61	9.82	8.9	9.81	9.45
Oxygen Saturation - OS	every 3 months	% O_2								
COD	every 3 months	mg/l	0.48	0.48	0.72	0.64	0.6	1.72	0.6	1.4
BOD_5	every 3 months	mg O_2/l	2.38	2.89	2.41	2.74	1.21	1.37	1.52	1.35
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.01	0.01	0.04	<0.001	0.03	0.04
NO_3-N	every 3 months	mg N/l	0.45	0.18	0.41	0.16	0.273	0.281	0.21	0.2
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	0.01	0.02
total nitrogen	every 3 months	mg N/l	0.18	0.22	0.2	0.21	0.309	0.353	0.27	0.25
ortho-phosphorus	every 3 months	µg P/l	10	10	20	20	4	4	10	10
TC	every 3 months	mg/l	2.17	2.49	3.5	3.61	3.87	10.2	3.29	3.71
Chloride	every 3 months	mg/l	3.55	3.55	5.33	3.55	2.01	2.198	2.14	2.18
Sulphate	every 3 months	mg/l	6.58	6.58	7.65	2.06	2.286	2.383	2.62	2.71
Chlorophyll a	only upper layer	40 µg/l (150 µg/l)	0.19				1.2			
Lead	one per year	mg/kg	<0.01	<0.01	<0.01		0.002		0.01	
Cadmium	one per year	mg/kg	0.008	<0.001	0.001		0.03		0.001	

Results for Lake Ohrid:

Station AL 1 – Lin

Parameters	Frequency	Unit of measurement	Surface	Surface	Surface	Surface
date of sampling			24/7/2013	8/10/2013	4/2/2014	13/04/2014
temperature	every 3 months	C	21	20	9.9	9.9
transparency	every 3 months	m	5	4.5	4	4
conductivity	every 3 months	µS/cm	231	229	234	236
pH	every 3 months		8.63	8.65	8.34	8.27
alkalinity as phenolphthalein (total alkalinity)	every 3 months	mg/l	118	105	113	112.5
suspended and dissolved inorganic and organic matter	every 3 months	mg/l	<0.1	<0.1	<0.1	<0.1
dissolved oxygen as mg×l-1 and oxygen saturation	every 3 months	% O_2	8.81	8.74	7.5	7.7
COD	if relevant impact of waste water - every 3 months	mg/l	0.4	0.48	0.32	0.44
BOD_5	every 3 months	mg O_2/l	1.02	0.48	1.67	1.67
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.02	0.02
NO_3-N	every 3 months	mg N/l	1.8	1.14	0.04	0.03
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	0.01	0.02
total nitrogen	every 3 months	mg N/l	1.7	1.5	0.127	0.14
total phosphorus	every 3 months	µg P/l	20	10	10	16
Chloride	every 3 months	mg/l	5.32	5.33	3.08	3.25
Sulphate	every 3 months	mg/l	9.88	4.94	6.762	6.85
Chlorophyll a	only upper layer	40 µg/l (150 µg/l)	1.91	0.92	0.594	
Lead	one per year	mg/kg			<0.01	0.01
Cadmium	one per year	mg/kg			<0.001	0.001
TC	every 3 months - only upper layer	mg/l	1.68	3.45	3.04	2.93

Station AL 2 – Memelishti

Parameters	Frequency	Unit of measurement	Surface	Surface	Surface	Surface
date of sampling			24/7/2013	8/10/2013	4/2/2014	13/04/2014
temperature	every 3 months	C	25.4	17.7	11.3	11.3
transparency	every 3 months	m	5	5.2	8	6
conductivity	every 3 months	µS/cm	236	230	232	236
pH	every 3 months		8.59	8.74	8.36	8.3
alkalinity as phenolphthalein (total alkalinity)	every 3 months	mg/l	118	110	113	112.5
suspended and dissolved inorganic and organic matter	every 3 months	mg/l	<0.01	<0.01	<0.01	<0.10
dissolved oxygen as mg×l-1 and oxygen saturation	every 3 months	% O_2	8.2	9.83	10.3	10.62
COD	if relevant impact of waste water - every 3 months	mg/l	0.4	0.56	0.32	0.32
BOD_5	every 3 months	mg O_2/l	1.25	0.52	2.02	2.02
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.01	0.01
NO_3-N	every 3 months	mg N/l	0.18	0.17	0.05	0.03
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	0.01	0.02
total nitrogen	every 3 months	mg N/l	0.31	0.13	0.205	0.13
total phosphorus	every 3 months	µg P/l	20	20	10	16
Chloride (parameter for salinity)	every 3 months	mg/l	5.32	3.55	3.08	3.12
Sulphate	every 3 months	mg/l	7.41	6.17	6.8	6.58
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	0.467	0.699	0.643	
Lead	one per year	mg/kg			<0.01	0.01
Cadmium	one per year	mg/kg			<0.001	0.001
TC	every 3 months - only upper layer	mg/l	0.31	3.34	3.55	0.13

Station AL 3 – Pogradeci (Tushemishti)

Parameters	Frequency	Unit of measurement	Surface	Surface	Surface	Surface
date of sampling			24/7/2013	8/10/2013	4/2/2014	13/04/2014
temperature	every 3 months	C	25	17.6	14.2	14.4
transparency	every 3 months	m	6	4.3	4	4
conductivity	every 3 months	µS/cm	235	232	234	241
pH	every 3 months		8.56	8.73	8.35	8.28
alkalinity as phenolphthalein (total alkalinity)	every 3 months	mg/l	120	110	113	115
suspended and dissolved inorganic and organic matter	every 3 months	mg/l	7	<0.10	<0.10	<0.1
dissolved oxygen as mg×l-1 and oxygen saturation	every 3 months	% O_2	8.1	9.48	7.4	6.48
COD	if relevant impact of waste water - every 3 months	mg/l	0.52	0.4	0.24	0.4
BOD_5	every 3 months	mg O_2/l	1.3	0.58	1.45	2.3
NH_3-N	every 3 months	mg N/l	<0.01	<0.01	0.02	0.02
NO_3-N	every 3 months	mg N/l	0.18	0.11	0.04	0.06
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	0.01	0.02
total nitrogen	every 3 months	mg N/l	0.41	0.14	0.147	0.19
total phosphorus	every 3 months	µg P/l	30	20	10	16
Chloride	every 3 months	mg/l	5.32	5.33	3.132	3.33
Sulphate	every 3 months	mg/l	10.29	7.41	6.939	6.85
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	0.478	0.643	0.923	
Lead	one per year	mg/kg			<0.01	0.01
Cadmium	one per year	mg/kg			<0.001	0.001
TC	every 3 months - only upper layer	mg/l	4.61	3.67	3.18	3.25

Results for Lake Prespa:

Station AL 1 – Gollomboc

Parameters	Frequency	Unit of measurement	Surface	Surface	Surface	Surface
date of sampling			24/7/2013	9/10/2013	5/2/2014	13/04/2014
temperature	every 3 months	C	27	18.2	5.13	11.1
transparency	every 3 months	m	3.5	3.5	3.5	4
conductivity	every 3 months	µS/cm	216	277	242	248
pH	every 3 months		8.77	8.43	8.06	8.37
alkalinity as phenolphthalein (total alkalinity)	every 3 months	mg/l	98	98	105	112.5
suspended and dissolved inorganic and organic matter	every 3 months	mg/l	3.7	0.5	<0.10	<0.10
dissolved oxygen as mg×l-1 and oxygen saturation	every 3 months	% O_2	7.5	12.4	5.74	14.63
COD	if relevant impact of waste water - every 3 months	mg/l	0.36	0.12	1.52	1.36
BOD_5	every 3 months	mg O_2/l	2.37	0.42	2.71	2.53
NH_3-N	every 3 months	mg N/l	<0.01	0.02	0.01	0.03
NO_3-N	every 3 months	mg N/l	0.14	0.09	0.01	0.02
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	<0.01	0.02
total nitrogen	every 3 months	mg N/l	0.45	0.39	0.338	0.33
total phosphorus	every 3 months	µg P/l	20	30	30	30
Chloride	every 3 months	mg/l	8.87	7.1	5.039	6.01
Sulphate	every 3 months	mg/l	12.76	7	10.119	10.55
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	2.29	5.116	2.952	1.412
Lead	one per year	mg/kg			<0.01	0.01
Cadmium	one per year	mg/kg			<0.001	0.001
TC	every 3 months - only upper layer	mg/l	4.64	6.04	5.57	5.43

Station AL 2 – Liqenas (Pusteci)

Parameters	Frequency	Unit of measurement	Surface	Surface	Surface	Surface
date of sampling			24/7/2013	9/10/2013	5/2/2014	13/04/2014
temperature	every 3 months	C	27	17.9	5.29	11.3
transparency	every 3 months	m	3.5	3.5	2.5	4
conductivity	every 3 months	µS/cm	209	225	241	248
pH	every 3 months		8.62	8.54	8.29	8.24
free carbon dioxide	every 3 months	ml HCl				
alkalinity as phenolphthalein (total alkalinity)	every 3 months	mg/l	93	98	110	112.5
suspended and dissolved inorganic and organic matter	every 3 months	mg/l	5	<0.01	<0.01	<0.10
dissolved oxygen as mg×l-1 and oxygen saturation	every 3 months	% O_2	7.71			11.26
COD	if relevant impact of waste water - every 3 months	mg/l	1.8	1.12	1.2	13.2
BOD_5	every 3 months	mg O_2/l	1.92	0.38	0.03	5.6
NH_3-N	every 3 months	mg N/l	<0.01	0.01	0.02	0.05
NO_3-N	every 3 months	mg N/l	0.72	0.74		0.02
NO_2-N	every 3 months	mg N/l	<0.01	<0.01	<0.01	0.02
$TN_{Kjeldahl\ nitrogen\ (organic\ nitrogen)}$	every 3 months	µg N/l				
total nitrogen	every 3 months	mg N/l	0.46	0.83	0.36	0.33
total phosphorus	every 3 months	µg P/l	20	30	30	23
Chloride	every 3 months	mg/l	7.1	8.88	4.995	5.58
Sulphate	every 3 months	mg/l	24.28	9.05	10.311	10.63
Chlorophyll a (relevant for bathing)	only upper layer	40 µg/l (150 µg/l)	3.226	5.811	2.874	1.633
Lead	one per year	mg/kg			<0.001	
Cadmium	one per year	mg/kg	<0.001		<0.001	
TC	every 3 months - only upper layer	mg/l		5.95	5.84	5.55

5. Interpretation

Lakes are not only valuable environmental resources. Furthermore economic (fishery, tourism, agriculture etc.) uses in the watershed could cause harmful impacts to the water quality and the ecosystem biodiversity. The results of the investigations are used for prelimary assessment of the trophic-status of the lakes .The results that are presented in the section 5 are assessed in this section to classify the waters of three lakes according to the WFD and trophic status indexes that are presented in the tables below:

Parameters	Unit of measurement	Threshold values according to the determination of trophic-status		
		Oligotrophic	Mesotrophic	Eutrophic
Transparency	m	5-10 (max 15-20)	1-2 (max.5-10)	<1 (max 2-3)
COD	mg/l	1-2	8-9	20-65
BOD_5	mg O_2/l	<3	3-5,5	5,5-14
NO_3-N	mg N/l	<1	<1	>2
Total phosphorus	µg P/l	4-10	10-35	35-100

Table 2. Natural water quality that support fish growing *(EC Decision: 78/659, dt. 18.07.1978; BMZ, 1995)*

Parameter	Salmonide water		Cyprinid waters	
	Mandatory level	Recommended level	Mandatory level	Recommended level
Temperature (ºC)	1. temperature measured at the edge of a mixture of thermal flow temperature should not be increased by			
	1.5ºC		3ºC	
	2. Thermal discharges must not cause an increase in temperature of the mixture over the limit:			
	21.5ºC		28ºC	
Dissolved oxygen (mg/l)	50% > 9	50% > 9 100% > 7	50% > 7	50% > 8 100% > 5
pH	6-9		6-9	
Total suspended solid (TSS, mg/l)	< 50	< 25	< 50	< 25
BOD_5		< 3		< 6
Orthophosphate (PO_4 mg/l)	0.2		0.4	
Nitrite (mg/l NO_2)		< 0.01		< 0.03
Ammonia (mg/l NH_4)	< 1	< 0.04	<1	< 0.2
NH_3 (mg/l)	< 0.025	< 0.005	< 0.025	< 0.005
Cl_2 (mg/l)	< 0.005		< 0.005	
Zinc total (mg/l Zn)	< 0.3		< 0.1	
Dissolved copper (mg/l Cu)		< 0.04		< 0.04

5.1 Lake Shkodra

Kalldrun (Bajza) station

This station is placed on the east part of the lake and the coast of the lake is flat and surrounded by the agriculture areas. The shore is flat and impacted from the flooding during rainy season.

The variation of the temperature (fig. 1) is depending from the season and the water flow. It varies from 18.1°C to 28.1°C in surface and from 14.1°C to 16.2°C in depth. Shkodra Lake is a shallow lake and the water circulate fast during the year and make homogenous layers for temperature and other water quality parameters such as pH (fig 2) (from 7.71 to 8.29) and conductivity (fig. 3) (from 221 ms/cm to 282 ms/cm).

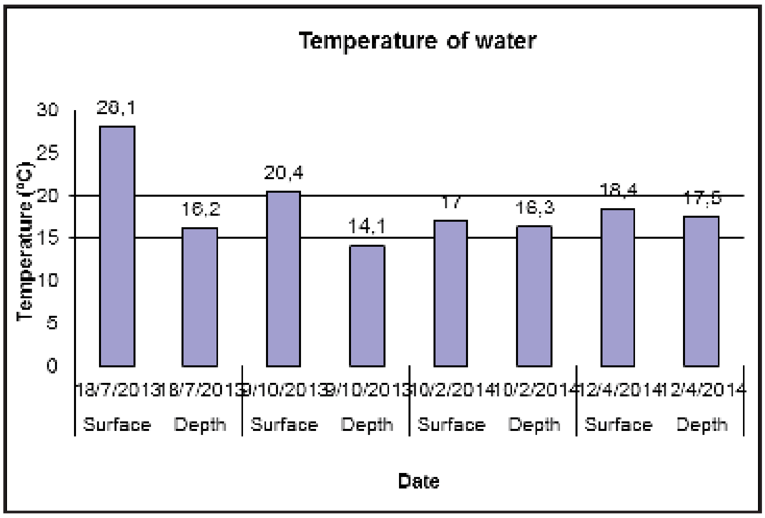


Figure 1. The variation of temperature at Kalldruni station

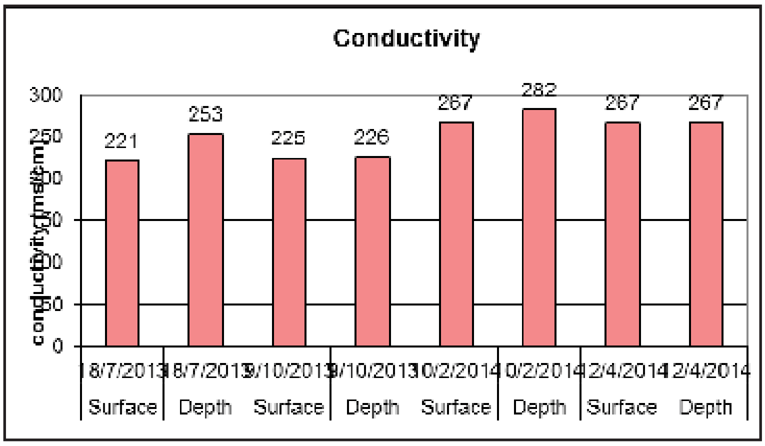


Figure 2. Variation of conductivity at Kalldruni station

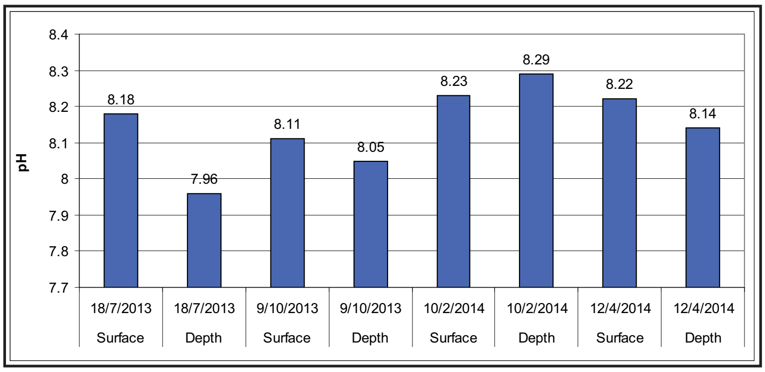


Figure 3. Variation of pH at Kalldruni station

The ***pH*** variation shows very well the mixing of the water during the different seasons where the difference between the surface and depth are small.

Alkalinity (fig.4)is a measure of the capacity of water to neutralize acids. Alkaline compounds in the water such as bicarbonates, carbonates, and hydroxides remove H^+ ions and decrease the acidity of the water (which means increased pH). To maintain a fairly constant pH in a water body, a higher alkalinity is preferable. High alkalinity means that the water body has the ability to neutralize acidic pollution from rainfall or basic inputs from wastewater. The values of this parameter at this station have no significant differences in different season and in water column. It varies from 113 mg/l in autumn (October) to 140mg/l in winter (February). So it seems that the water has high neutralize capacity of acidic pollution that come from inland waters and discharge into lake.

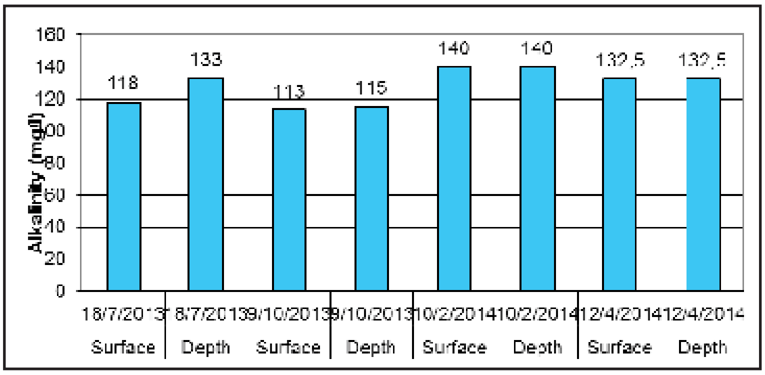


Figure 4. Variation of alkalinity content in water at Kalldruni station

Results are showing the mixing of the waters and no differences in values of ***DO***. When the waters have high temperatures the amount of DO is low (fig. 5). DO doesn`t show greater differences over the year.

The measured low values (red ones) are not correct and it depends from the calibration of the field equipment.

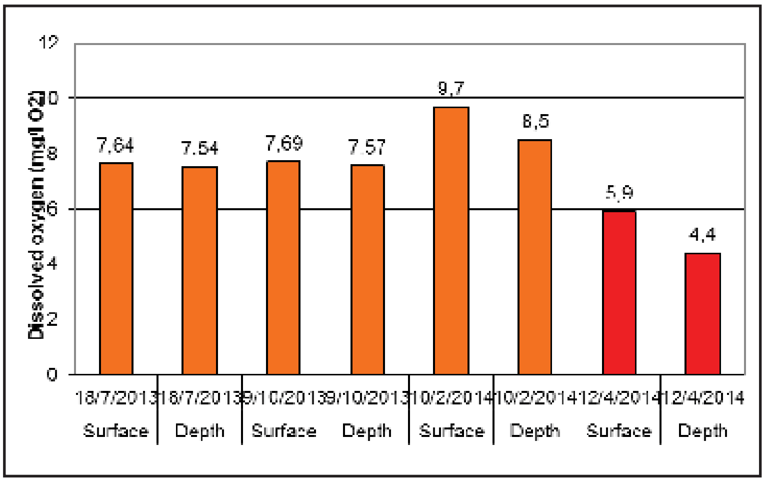


Figure 5. Variation of the dissolved oxygen at Kalldruni station

The same results are taken from the ***TC and BOD*** content in waters at Kalldruni station (fig.6). The values have variation that depends from the temperature of water and also the biological activities in different seasons.

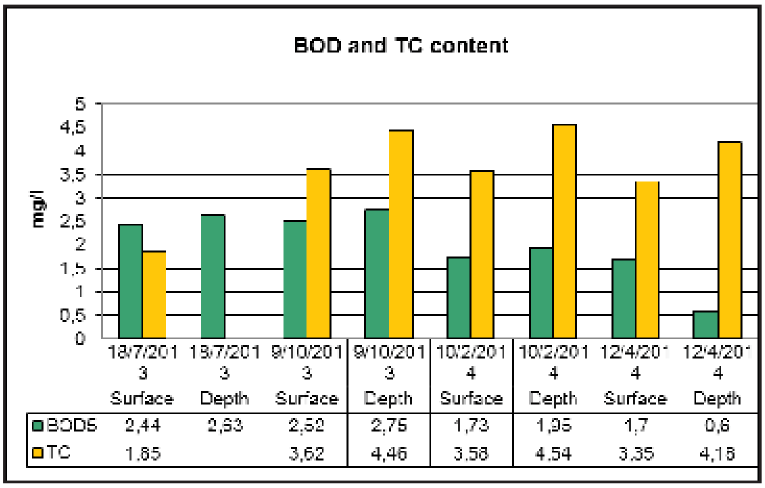


Figure 6. BOD and TC content at Kalldruni station

Values of BOD and TC show approximately the same level of them in ratio that means the same level of pollution impact in different seasons. The values of BOD are at typical level of pollution that comes from the urban and agriculture discharges. The ratio BOD/TC is at the low level that means the inorganic pollution is at low level.

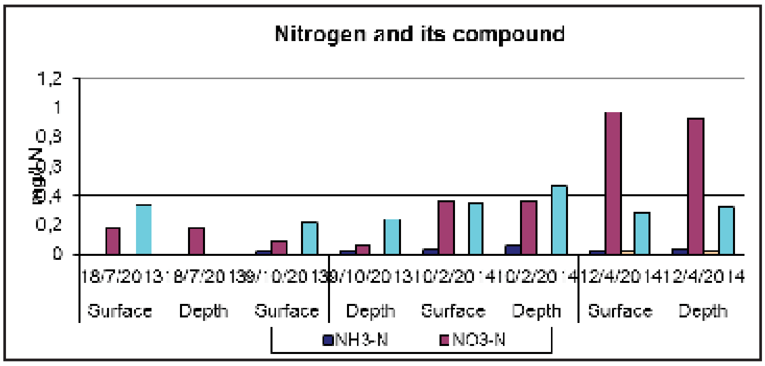


Figure. 7 Nitrogen and its compound content at Kalldruni station

Nitrogen content is not high at this station even there is an agriculture impact that comes from the fertilizers used in arable areas and splashed from them during the rainy period. Values of nitrates are changing over the year (from 0.18 to 0.97 mg N/l). In the spring season the concentration is usally higher because of the use of fertilizers. The other compounds of nitrogen (ammonia from 0.02 to 0.06 mg N/l and nitrites from 0.02 to 0.14 mg N/l) are below the threshold level (< 1mg N/l).

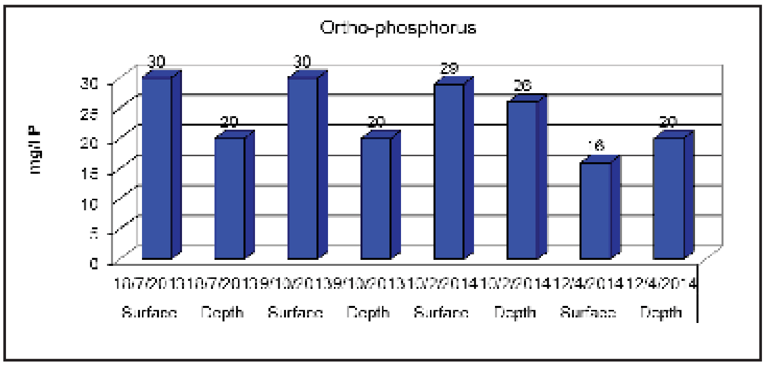


Figure 8. Phosphorous content at Kalldruni station

The phosphorous content (fig. 8) is at a mesotrophic level. High values (from 16 – 20 mg/l) of ortho phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Phosphorous intensives the eutrophication process and the risk of failing a good status according the WFD.

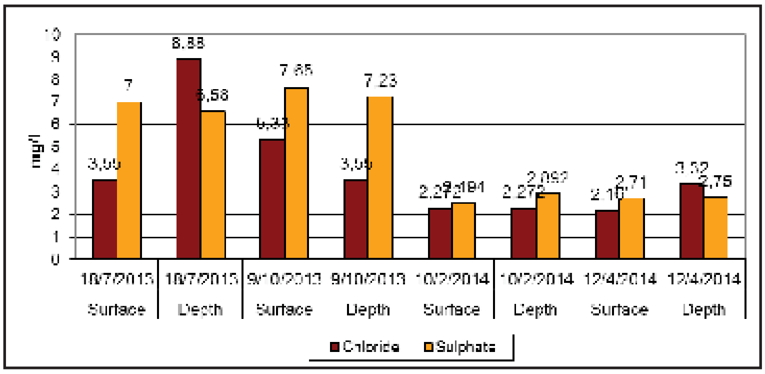


Figure 9. Sulphate and chloride content at Kalldruni station water

These parameters are not so significant at the assessment of the water quality but they play a role in relation of ions balance in water and express the impact from the ground of the lake. Values of these parameters are in normal level of the karstic environment of ground of the lake.

The trophic status of the water quality assess by parameters such as transparency (fig. 11), nutrients content (P and N) and the chlorophyll. All these parameters give an important contribution in assessment of the trophic level of waters. At Kalldruni station the values of chlorophyll a are taken in summer and autumn season when the biological activity is more developed. At the figure 10 is presented the values of this parameter in different season and can see the high presence of it in autumn (1.2 mg/l).

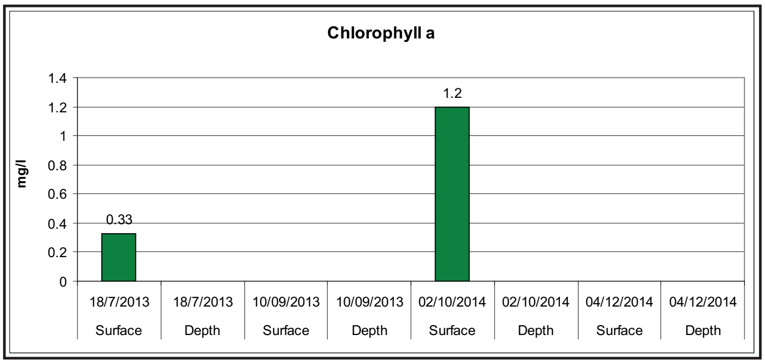


Figure 10. Chlorophyll a content at Kalldruni station

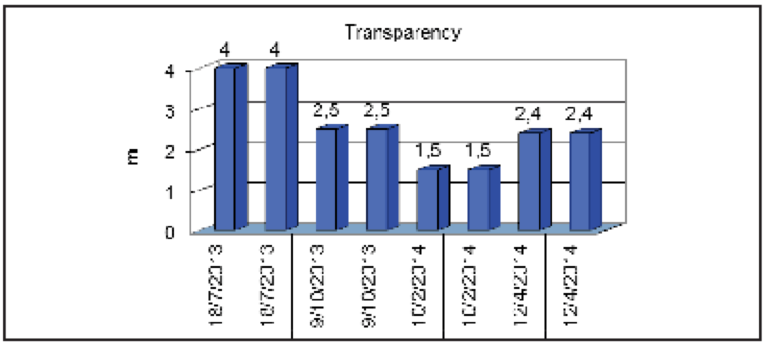


Figure 11. Transparency of the water at Kalldruni station

Zogaj station

This station is placed on the west part of the lake and the coast of the lake is gravel and rocky and there are agriculture and tourist areas.

The variation of the temperature (fig. 12) is depending from the season and the water flow. It varies from 18.3°C to 27.8°C. Shkodra Lake is a shallow lake and the water circulate fast during the year and make homogenous layers for temperature and other physical and chemical water quality parameters such as pH (fig 13) (from 8.26 to 8.46 pH unit) and conductivity (fig. 14) (from 222 ms/cm to 269 ms/cm). Values are at the same level with the other station in Shkodra Lake.

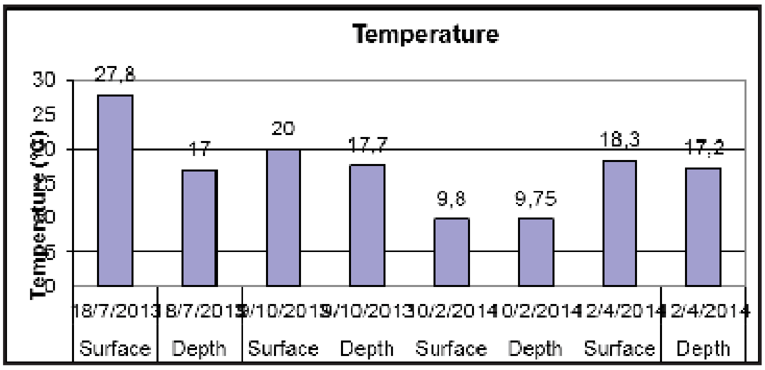


Figure 12. The variation of temperature at Zogaj station

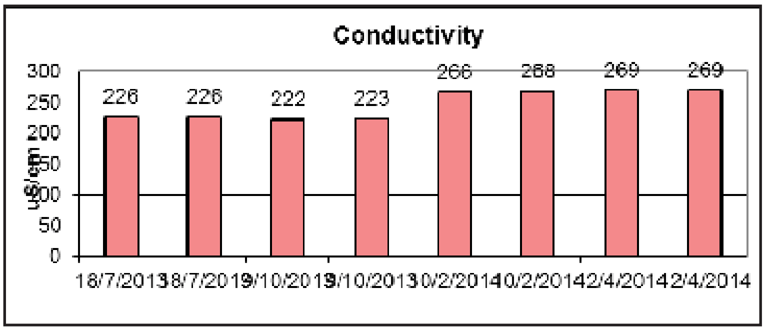


Figure 13. Variation of conductivity at Zogaj station

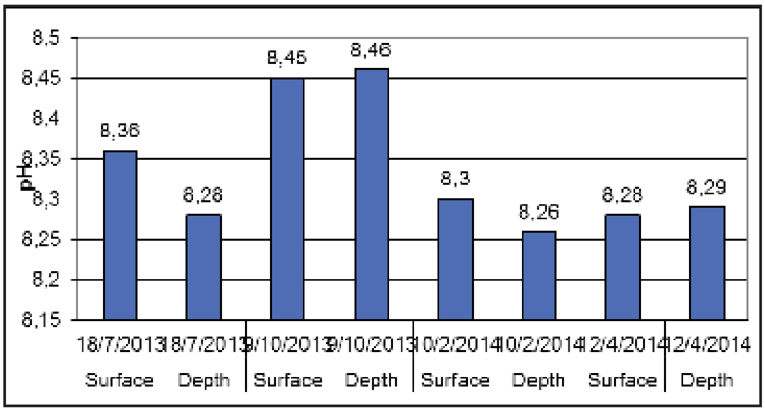


Figure 14. Variation of pH at Zogaj station

The same discussion we can make even for the ***alkalinity*** of the waters (fig. 15). The values of this parameter have no significant differences in different season and in water column. It varies from 113 mg/l in autumn (October) to 138 mg/l in winter (February).

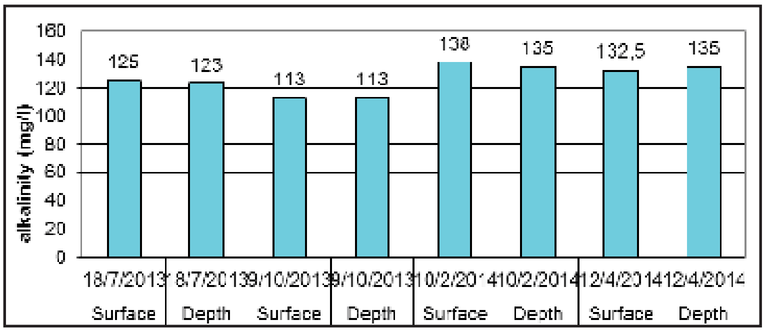


Figure 15. Variation of alkalinity content in water at Zogaj station

Water of lake shows in different season of the year that the DO content depending from the climate indication in water quality. When the temperature is low and the wind is present during the sampling campaign, the results showing the mixing of the waters and no differences in values of ***DO***. When the waters have high temperatures the amount of DO is low (fig. 16). According to the results we can see that in different seasons the DO content has no significant differences.

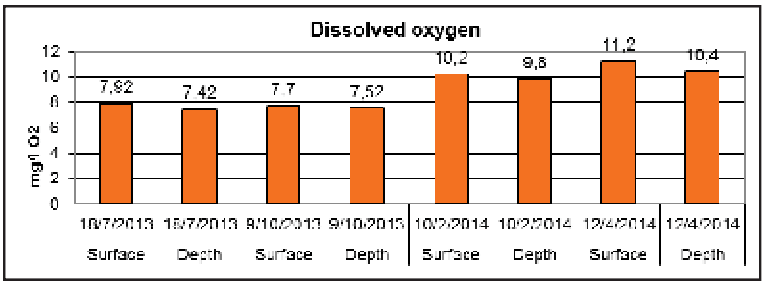


Figure 16. Variation of the dissolved oxygen at Zogaj station

The results taken for the ***BOD*** content in waters at Zogaj station (fig. 17) has not differences. The values have variation that depends from the temperature of water and also the biological activities in different seasons. For the BOD vary from 1.3 to 2.24 mg/l O_2.

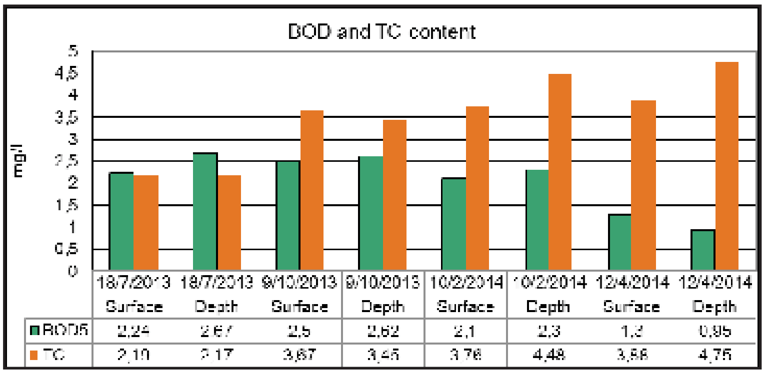


Figure 17. TC and BOD content at Zogaj station

The values of BOD are lower than the threshold level (< 3 mg/l O_2) so the biological impact from the waste water discharges is very low.

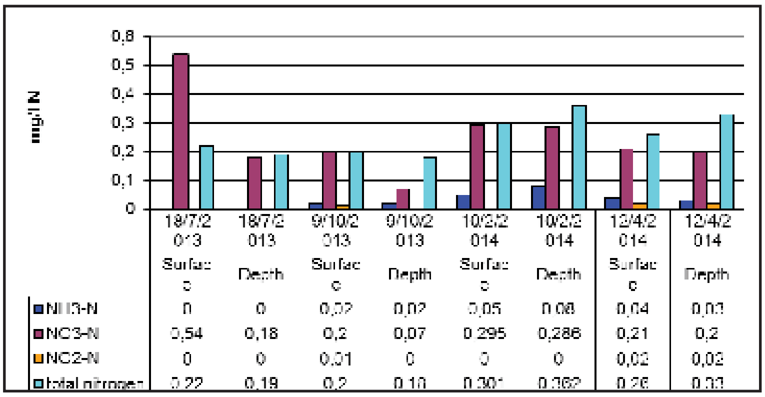


Figure. 18 Nitrogen and its compound content at Zogaj station

Nitrogen content is not high at this station. As we can see the same period of year shows higher values of nitrates content (from 0.07 to 0.82 mg N/l) and it is the spring season when the agriculture activities are more developed. The other compounds of nitrogen (ammonia from 0.02 to 0.08 mg N/l and nitrites from 0.01 to 0.13 mg N/l) are below the threshold level (< 1).

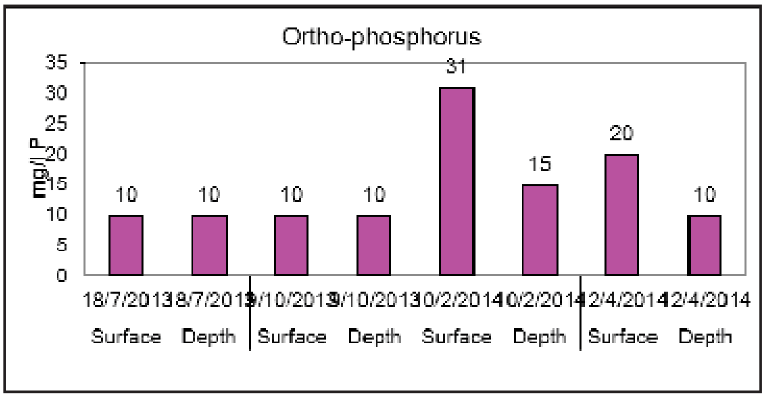


Figure 19. Phosphorous content at Zogaj station

Phosphorous (fig. 19) concentration is at mesotrophic level. High values (from 10 – 20 mg/l) of ortho-phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Generally values at this level are not in accordance for a good ecological water status.

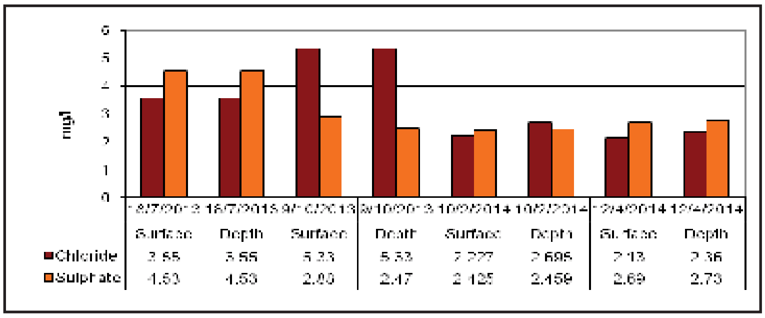


Figure 20. Sulphate and chloride content at Zogaj station water

These parameters are in normal level of the karstic environment of ground of the lake. The trophic status of the water quality assess by parameters such as transparency (fig. 22), nutrients content (P and N) and the chlorophyll a (fig. 21). All these parameters give an important contribution in assessment of the trophic level of waters. At the figure 21 is presented the values of this parameter in different season and can see the high presence of it in autumn .

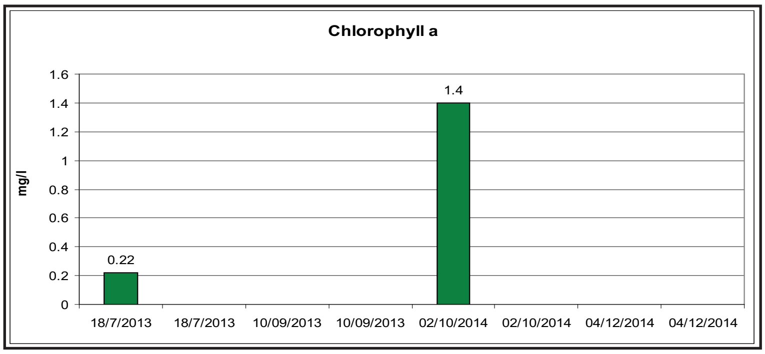


Figure 21. Chlorophyll a content at Zogaj station

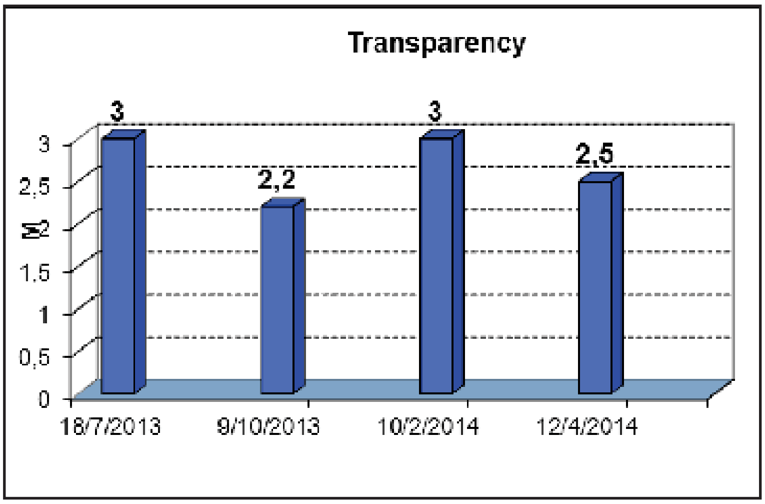


Figure 22. Transparency at Zogaj station

Shiroka station

This station is placed on the southwest part of the lake and the coast of the lake is flat combined with gavel and surrounded by the small agriculture and tourist areas.

The variation of the temperature (fig. 23) is depending from the season and the water flow. It varies from 17.9°C to 28.3°C. As we can see the temperatures at this station are higher than other. The homogeneity layers for temperature and other water quality parameters such as pH (fig 24) (from 7.71 to 8.29) and conductivity (fig. 25) (from 221 ms/cm to 282 ms/cm) are observed even for this station.

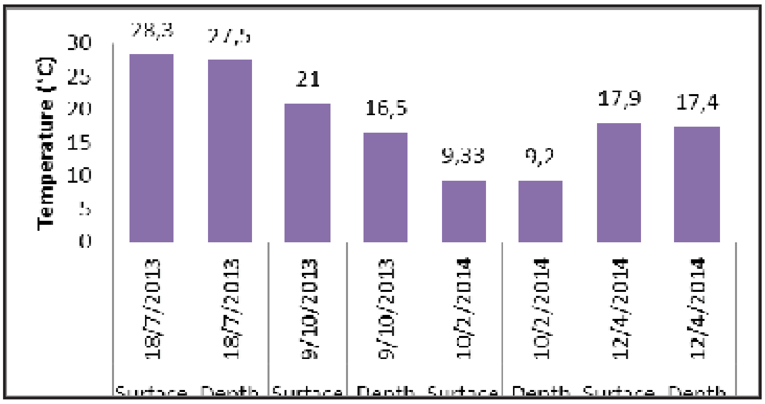


Figure 23. The variation of temperature at Shiroka station

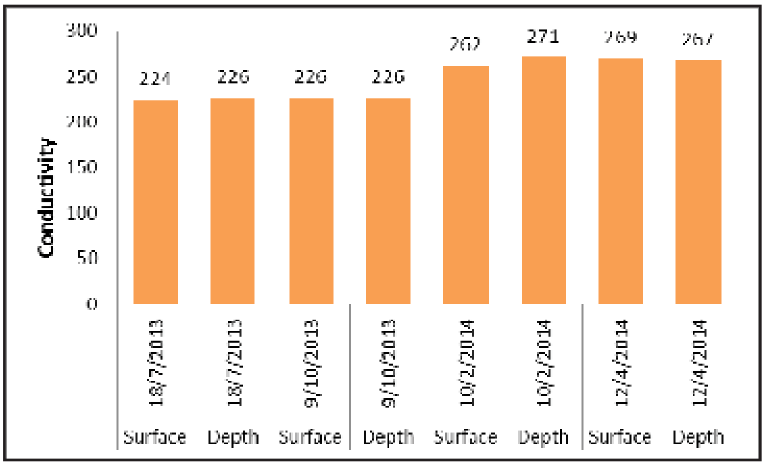


Figure 24. Variation of conductivity at Shiroka station

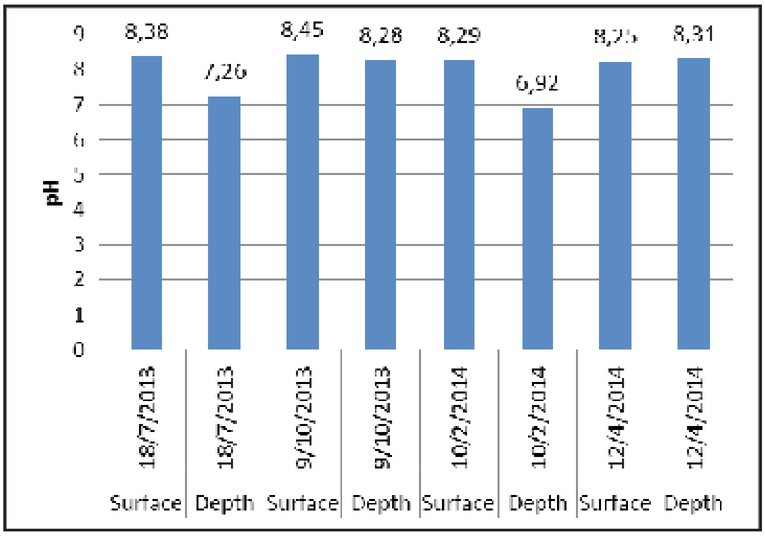


Figure 25. Variation of pH at Shiroka station

The values of ***pH*** show very well the mixing of the water during the different seasons where the difference between the surface and depth are small and from season to other they vary from 8.25 to 8.45.

The same interpretation we can make even for the ***alkalinity*** of the waters (fig.26). The values of this parameter vary from 114 mg/l in autumn (October) to 140 mg/l in winter (February).

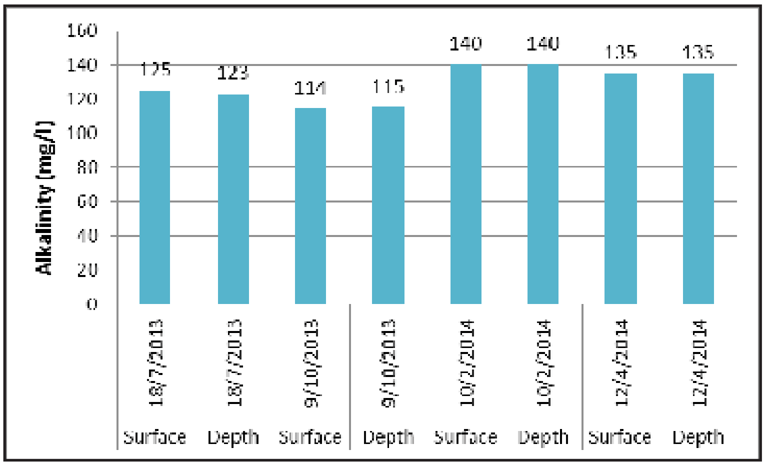


Figure 26. Variation of alkalinity content in water at Shiroka station

Dissolved oxygen shows different values depending on climate indication in water quality. When the temperature is low and the wind is present during the sampling campaign, the results showing the mixing of the waters and small differences of ***DO values from surface to depth***. When the waters have high temperatures the amount of DO is low (fig. 27).

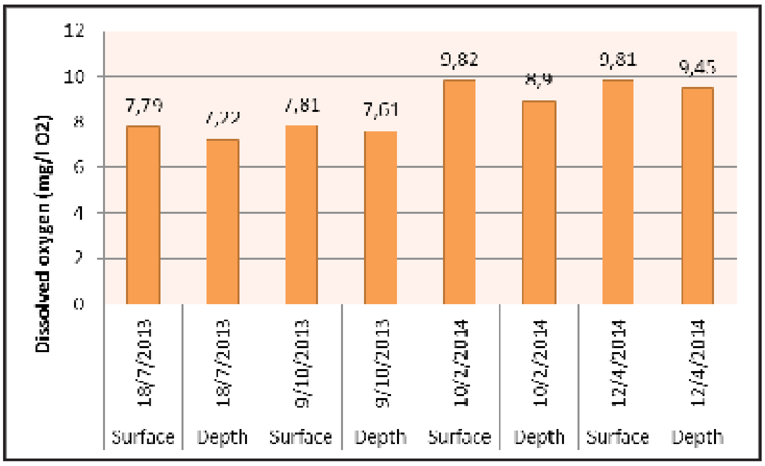


Figure 27. Variation of the dissolved oxygen at Shiroka station

The same results are taken from the ***BOD and TC*** content in waters at Shiroka station (fig.28). The values have variation that depends from the temperature of water and also the biological activities in different seasons.

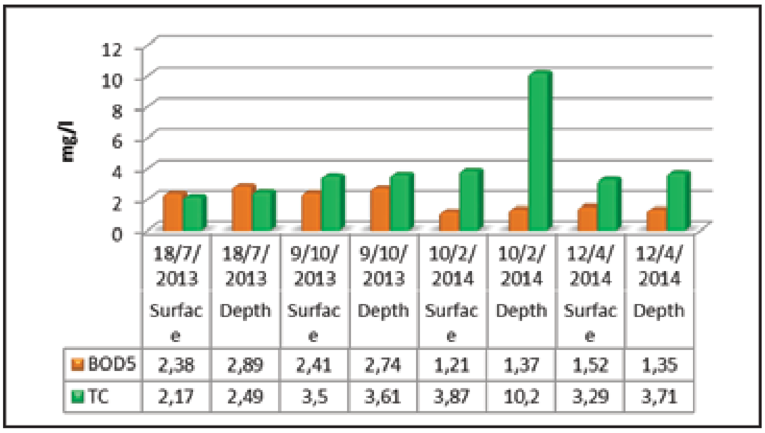


Figure 28. BOD and TC content at Shiroka station

The values of BOD are lower than the threshold level so the biological impact from the waste water discharges is very low. They vary from 1.52 mg/l O_2 in spring to 2.38 mg/l O_2 in summer.

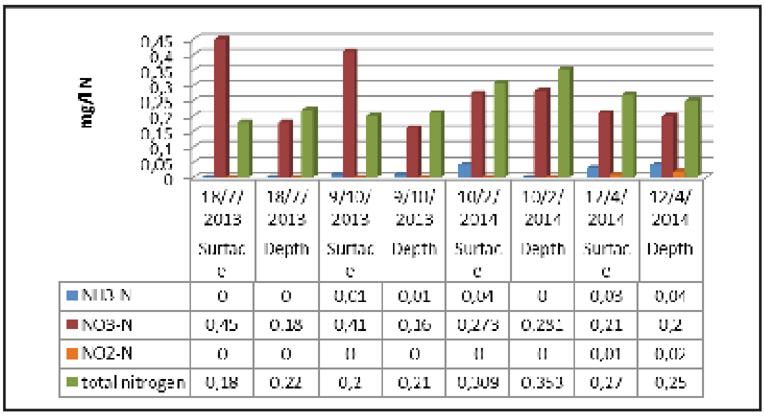


Figure. 29. Nitrogen and its compound content at Shiroka station

Nitrogen content is not high at this station (fig. 29). As we can see along the year it shows approximately the same values of nitrates content from 0.16 to 0.2 mg N/l at the bottom and from 0.21 to 0.45 mg N/l at the surface. The other compounds of nitrogen (ammonia from 0.01 to 0.04 mg N/l and nitrites from 0.01 to 0.02 mg N/l) are below the threshold level (< 1).

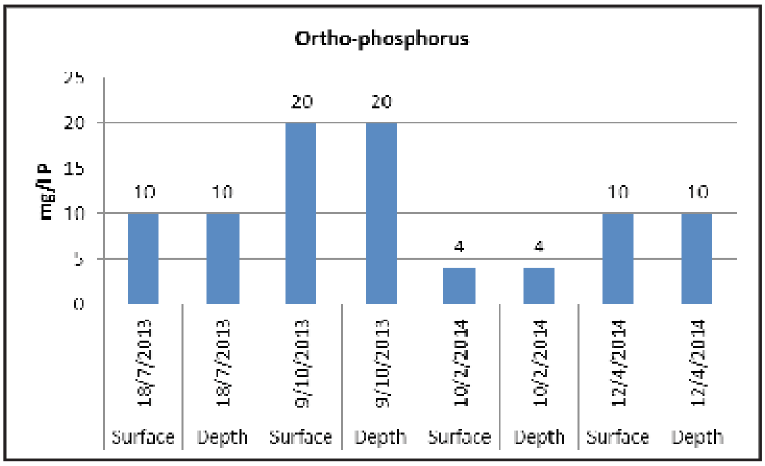


Figure 30. Phosphorous content at Shiroka station

The phosphorous content (fig. 30) is at mesotrophic level. The values (from 10 – 20 mg/l) of ortho phosphorous indicate risks in water quality as a limited factor of biological development of water life.

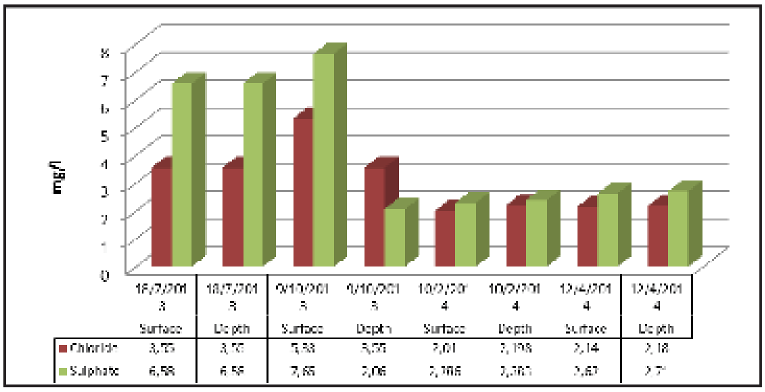


Figure 31. Sulphate and chloride content at Shiroka station water

These parameters are not so significant at the assessment of the water quality but they play a role in relation of ions balance in water and express the impact from the ground of the lake. Values of these parameters are in normal level (fig. 31).

The trophic status of the water quality assesses by parameters such as transparency (fig. 33), nutrients content (P and N) and the chlorophyll a (fig.32). All these parameters give an important contribution in assessment of the trophic level of waters. At Shiroka station the values of chlorophyll a are taken in summer and winter season. At the figure 32 is presented the values of this parameter in different season and can see the high presence of it in winter (1.2 mg/l).

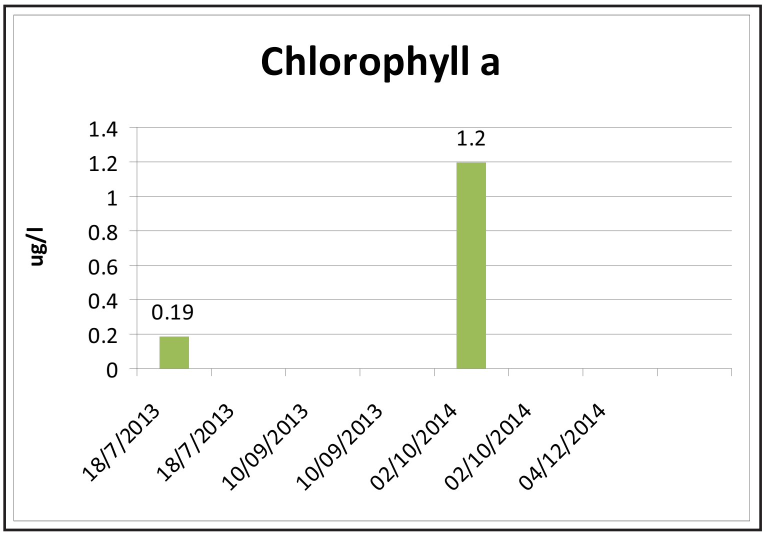


Figure 32. Chlorophyll a content at Shiroka station

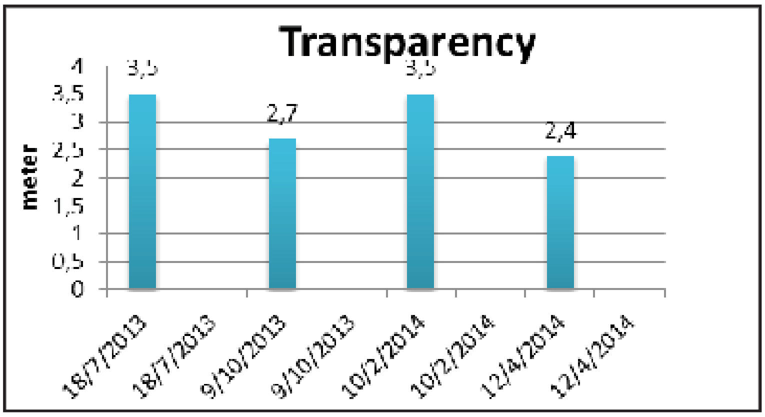


Figure 33. Transparency of water at Shiroka station

The classification of Shkodra Lake water made by using the Carlson Trophic State Index as prescribed below:

Trophic Class	Chlorophyll a content (mg/l)	Phosphorous content (mg/l).	Secchi disc (m)	Trophy Index
Oligotrophic	0—2.6	0—12	>8—4	<30—40
Mesotrophic	2.6—20	12—24	4—2	40—50
Eutrophic	20—56	24—96	2—0.5	50—70
Hypereutrophic	56—155+	96—384+	0.5—<0.25	70—100+

a. TSI for Chlorophyll-a (CA) TSI = 9.81In Chlorophyll-a(ug/L)+30.6

b. TSI for Secchi depth (SD) TSI = 60-14.41In Secchi depth(Meters)

c. TSI for Total phosphorous (TP) TSI = 14.42 In Total phosphorous (ug/l) + 4.15

where TSI is Carlson Trophic State Index and In is Natural logarithm

Carlson's TSI = [**TSI (TP)**+**TSI(CA)**+**TSI(SD)**] / **3**

where TP and Chlorophyll-a in micrograms/l and SD transparency in meters.

After the calculation of each index (table below):

Station	Chlorophyll a content (mg/l)	TSI (CA)	Secchi disc (m)	TSI (SD)	Phosphorous content (mg/l).	TSI (TP)	Trophy Index
Kalldrun (Bajza)	0.33	20	4	40	10	37	32
			4	40	10	37	26
			2.5	47	10	37	28
			2.5	47	10	37	28
	1.2	32	1.5	54	10	37	41
			1.5	54	10	37	31
			2.4	47	16	44	31
			2.4	47	20	47	32
Zogaj	0.22	16	3	44	10	37	27
					10	37	12
			2.2	49	10	37	29
					10	37	24
	1.4	34	3	44	10	37	38
					10	37	16
			2.5	47	20	47	28
					10	37	12
Shiroka	0.19	14	3.5	42	10	37	26
					10	37	16
			2.7	46	20	47	31
					20	47	8
	1.2	32	3.5	42	4	24	33
					4	24	8
			2.4	47	10	37	28
					10	37	12

Carlson Trophic State Index :

- *Kalldruni station* - mesotrophic state for Secchi disc and Chlorophyll and oligotrophic state for TSI
- *Zogaj station* - mesotrophic state for Secchi disc and Chlorophyll and oligotrophic state for TSI
- *Shiroka station* - mesotrophic state for Secchi disc and Chlorophyll and oligotrophic state for TS

According the determination of the ecological status of the water bodies physico-chemical elements have only a supporting character.

5.2 Lake Ohrid

There are three stations in Ohrid Lake involved in monitoring program and for evaluation of the lake water status. The result interpretation will be made station by station and an overall water classification will be discussed for the lake.

Lin station

This station is placed on the northwest part of the lake and the coast of the lake is rocky with underwater hills and valleys.

Below we are interpreted the results taken from the analyses of the water samples taken in different seasons according to the monitoring program implementation.

The variation of the temperature (fig. 34) is depending from the season and varies from 9.9°C to 21°C. Ohrid Lake is a deep lake and the water didn't circulate fast during the year and the layers of water column have different temperature from the surface to the bottom of the lake. Also other water quality parameters such as pH (fig 35) (from 7.71 to 8.29) and conductivity (fig. 36) (from 221 ms/cm to 282 ms/cm) have different values during the year.

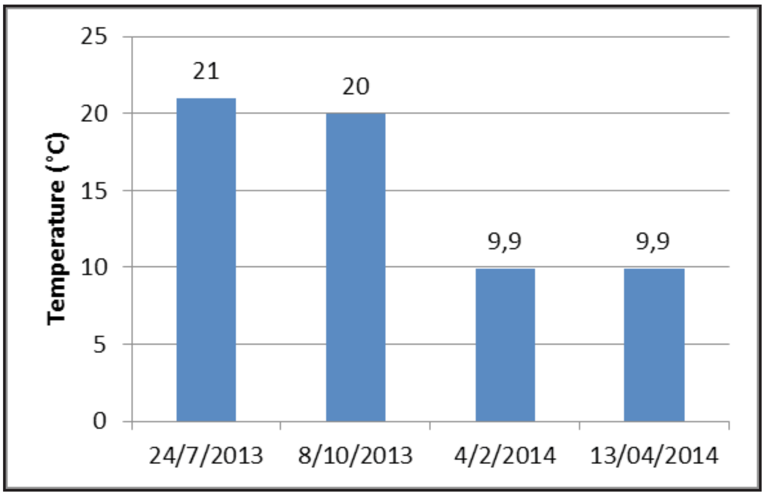


Figure 34. The variation of temperature at Lin station

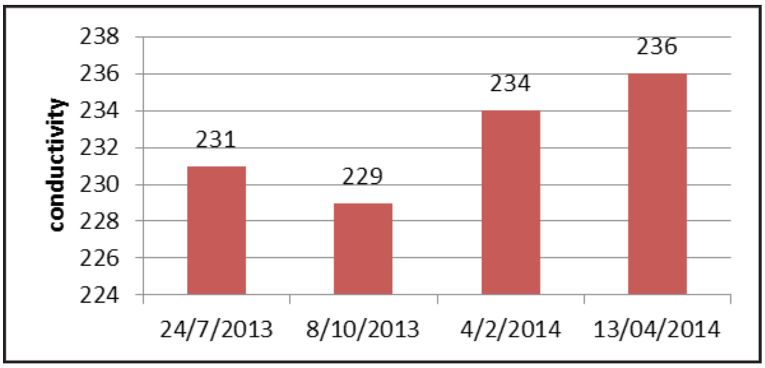


Figure 35. Variation of conductivity at Lin station

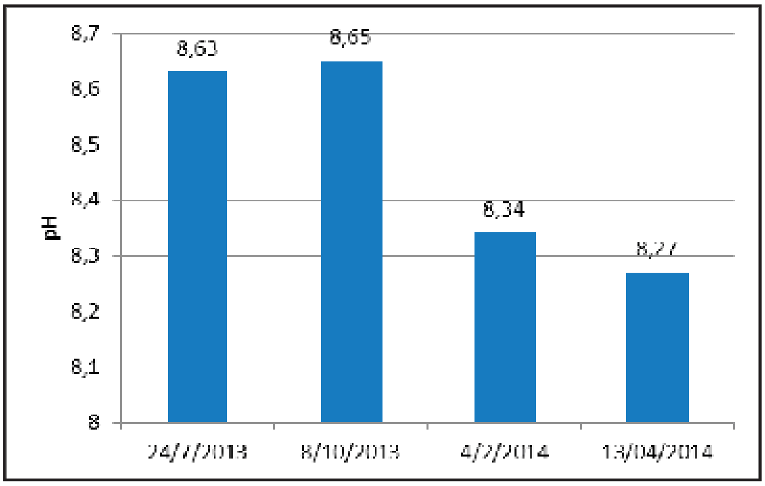


Figure 36. Variation of pH at Lin station

The ***pH*** variation shows very well the changes between the climate conditions during the year and they are depended by season temperature.

The same discussion we cannot make for the ***alkalinity*** of the waters (fig.37). The values of this parameter have no significant differences in different season and in water column. It varies from 105 mg/l in autumn (October) to 113 mg/l in winter (February).

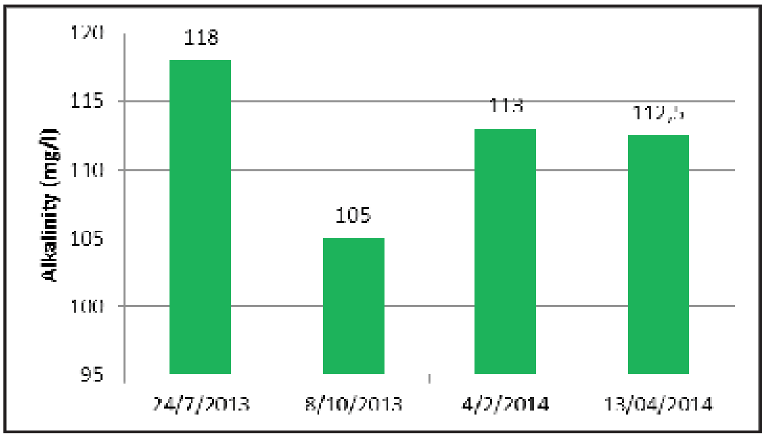


Figure 37. Variation of alkalinity content in water at Lin station

Water of lake is well oxygenated in different season of the year even there are some climate indication in water quality. When the temperature is low and the wind is present during the sampling campaign, the results showing high values of ***DO***. When the waters have high temperatures the amount of DO is low (fig. 38). According to the results we can see that in different seasons the DO content has differences.

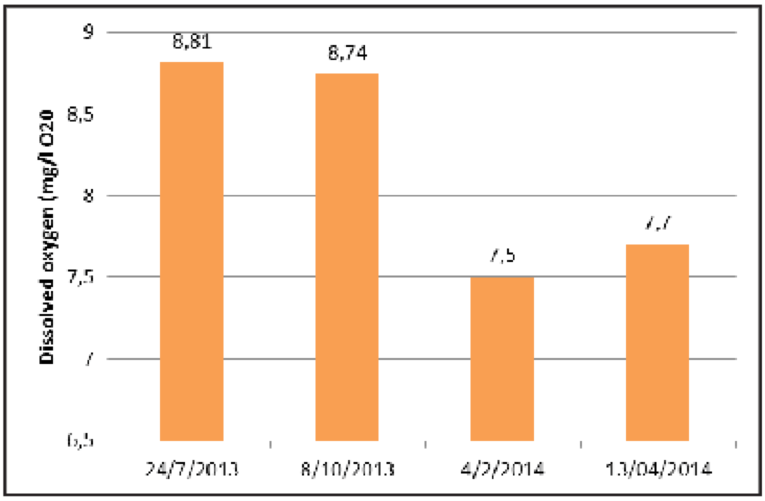
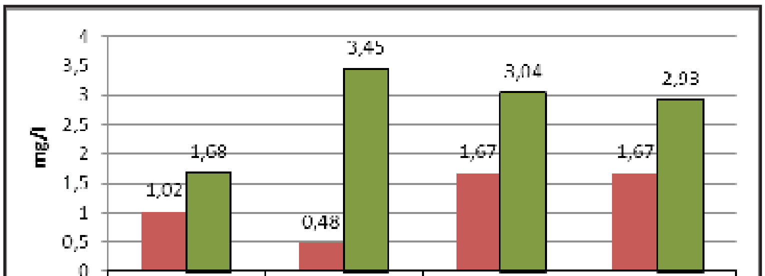


Figure 38. Variation of the dissolved oxygen at Lin station

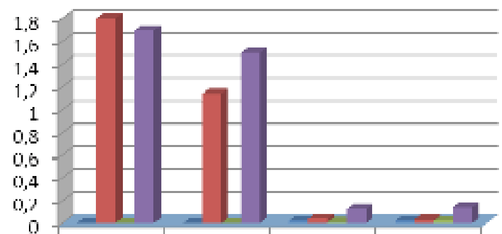
The results taken for ***TC and BOD*** content in waters at Lin station (fig.39) presented the different situation in different season. The values of BOD vary from 0.48 mg/l O_2 in summer to 1.67 mg/l O_2 in spring that shows the biological activities during these seasons.



	24/7/2013	8/10/2013	4/2/2014	13/04/2014
BOD5	1,02	0,48	1,67	1,67
TC	1,68	3,45	3,04	2,93

Figure 39. BOD and TC content at Lin station

The values of BOD are lower than the threshold level so the biological impact from the waste water discharges is very low.



	24/7/2013	8/10/2013	4/2/2014	13/04/2014
NH3-N	0	0	0,02	0,02
NO3-N	1,8	1,14	0,04	0,03
NO2-N	0	0	0,01	0,02
total nitrogen	1,7	1,5	0,127	0,14

Figure 40. Nitrogen and its compound content at Lin station

Nitrogen content is not high at this station (fig. 41). The values of nitrates content vary from 0.03 to 1.8 mg N/l at the spring and summer season. The other compounds of nitrogen like nitrites vary from 0.01 to 0.02mg N/l and they are below the threshold level (< 1).

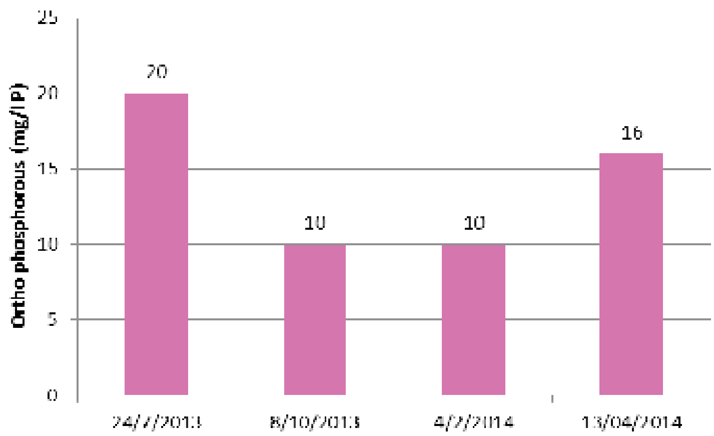
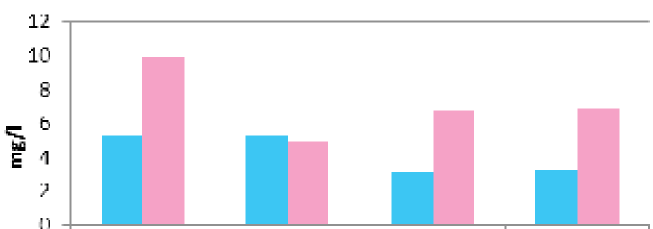


Figure 42. Phosphorous content at Lin station

The phosphorous content (fig. 42) is at mesotrophic level. The values (from 10 – 20 mg/l) of total phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Presence of phosphorous in this level has a negative impact for the good quality of shallow waters like coastal that are impacted from urban discharges.



	24/7/2013	8/10/2013	4/2/2014	13/04/2014
Chloride	5,32	5,33	3,08	3,25
Sulphate	9,88	4,94	6,762	6,85

Figure 43. Sulphate and chloride content at Lin station

These parameters are significant for the assessment of the water ions balance and express the impact from the ground of the lake. Values of these parameters are in normal level for the environment of ground of Ohrid Lake.

The trophic status of the water quality assess by parameters such as transparency (fig. 45), nutrients content (P and N) and the chlorophyll a (fig. 44). All these parameters give an important contribution in assessment of the trophic level of waters. At Lin station the values of chlorophyll a are taken in different seasons. At the figure 44 is presented the values of this parameter in different season and can see the high presence of it in summer and autumn (1.91 – 0.92 mg/l).

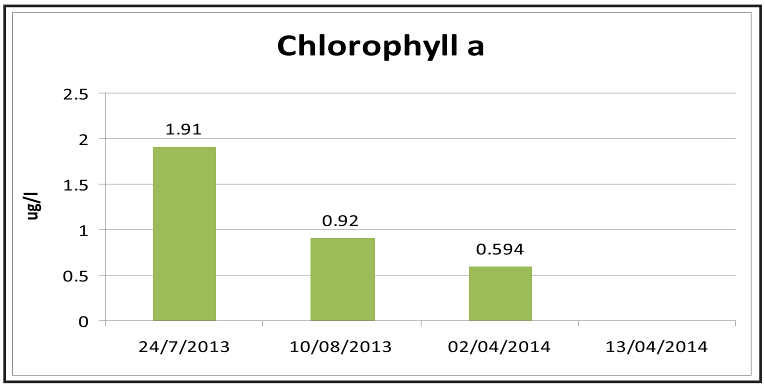


Figure 44. Chlorophyll a content at Lin station

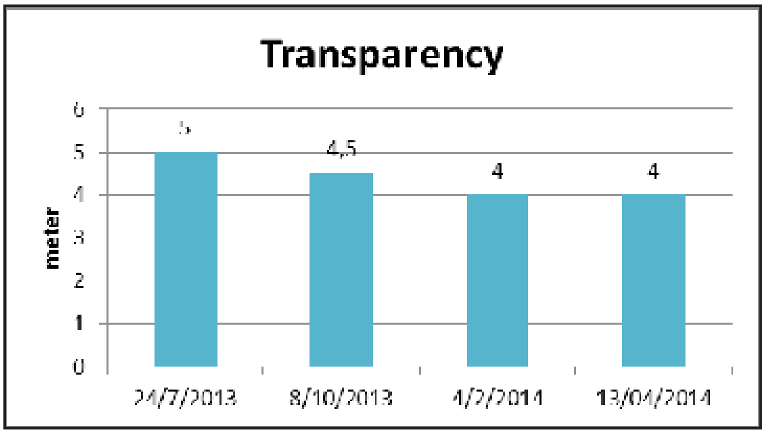


Figure 45. Transparency of the water at Lin station

Memelisht station

This station is placed on the west part of the lake and the coast of the lake is gravel and rocky. This part of the lake is impacted by the mineral damp and the mine stream flows.

The variation of the temperature (fig. 46) is depending from the season. It varies from 11.3°C to 25.4°C. The layers of water column have different temperature and other water quality parameters such as pH (fig 47) (from 7.89 to 8.46) and conductivity (fig. 48) (from 222 ms/cm to 269 ms/cm).

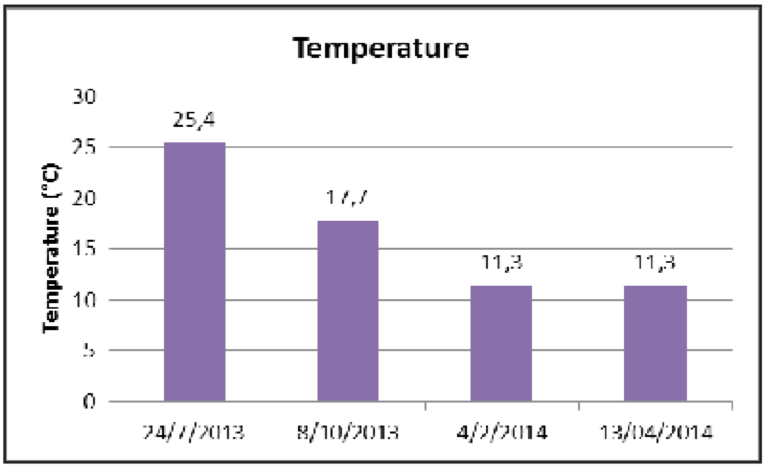


Figure 46. The variation of temperature at Memelishti station

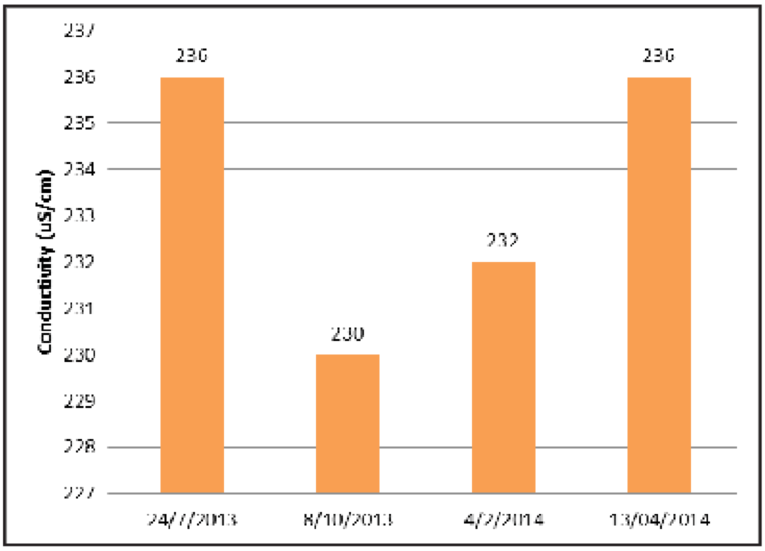


Figure 47. Variation of conductivity at Memelishti station

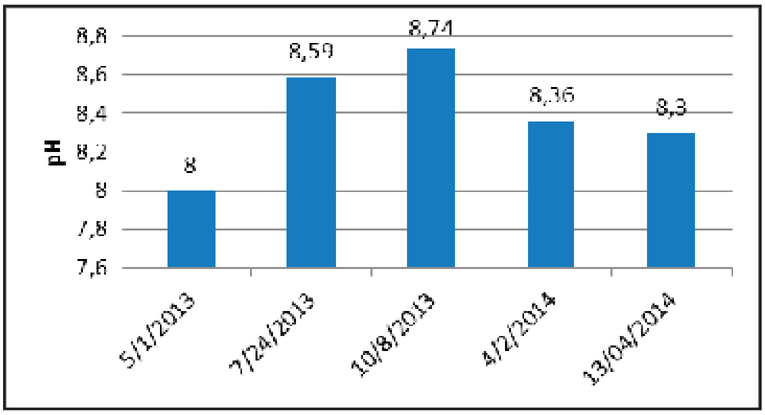


Figure 48. Variation of pH at Memelishti station

The same discussion we can make even for the ***alkalinity*** of the waters (fig.49). The values of this parameter have no significant differences in different season and in water column. It varies from 110 mg/l in autumn (October) to 118 mg/l in spring (July).

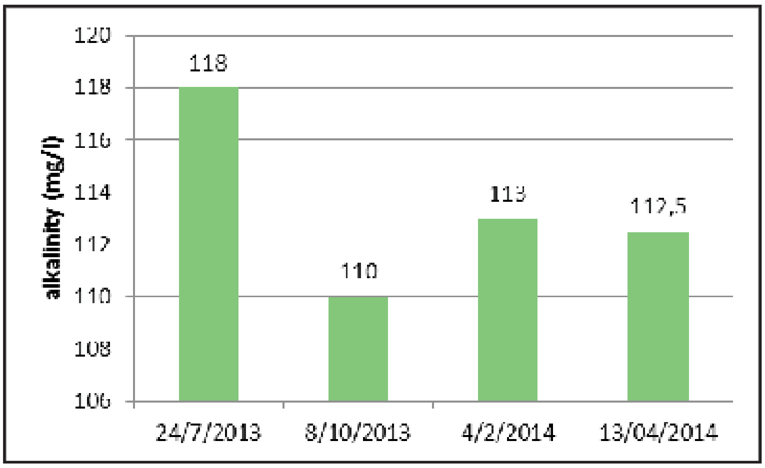


Figure 49. Variation of alkalinity content in water at Memelisht station

Results of DO content show they depended from the climate indication in water quality in different season of the year. When the waters have high temperatures the amount of DO is low (fig. 50). According to the results we can see that in different seasons the DO content has differences.

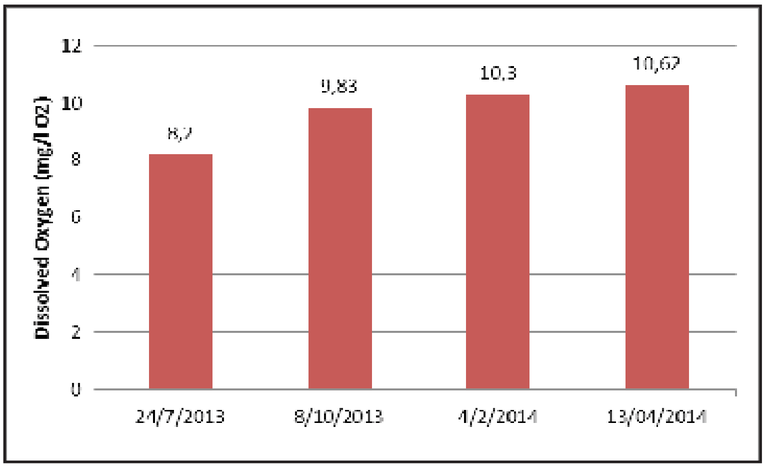


Figure 50. Variation of the dissolved oxygen at Memelishti station

The results of the ***TC and BOD*** content in waters at Memelishti station are shown in figure 51. The values have variation that depends from the temperature of water and also the biological activities in different seasons. For the TC values vary from 0.31 (in summer) to 3.55 mg/l O_2 (in winter) and for BOD vary from 0.52 (in autumn) to 2.02 mg/l O_2 (in spring).

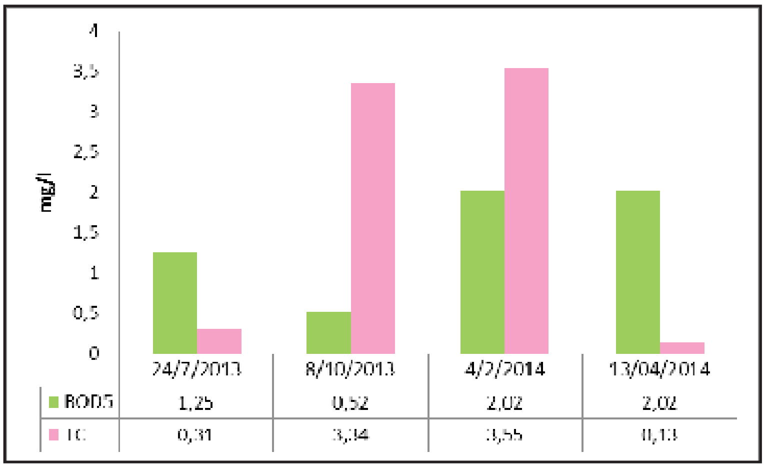


Figure 51. BOD and TC content at Memelishti station

The values of BOD are lower than the threshold level (< 3 mg/l O_2) so the biological impact from the waste water discharges is very low.

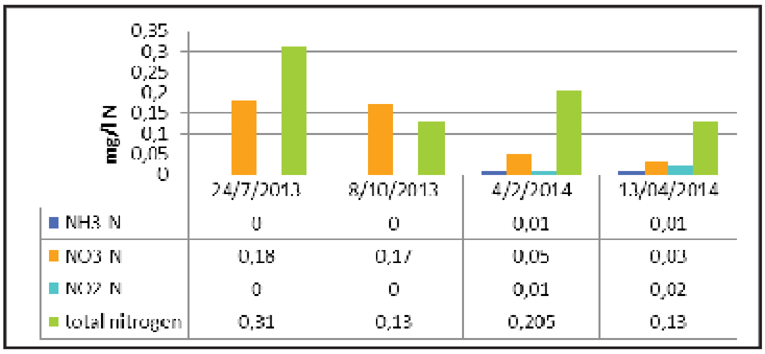


Figure 52. Nitrogen and its compound content at Memelishti station

Nitrogen content is not high even at this station. As we can see the values of nitrates content vary from 0.03 (spring) to 0.18 mg N/l (in summer). The other compounds of nitrogen ammonia are at the lowest level and nitrites vary from 0.01 to 0.02 mg N/l. They are below the threshold level (< 1).

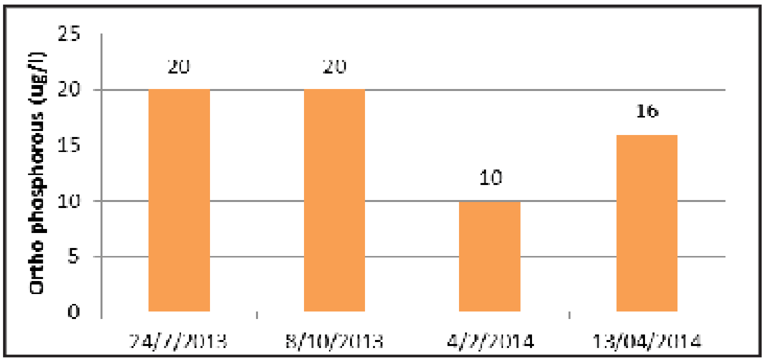


Figure 53. Phosphorous content at Memelishti station

The phosphorous content (fig. 53) is at mesotrophic level. High values (from 10 – 20 mg/l) of total phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Presences of phosphorous in this level is the same as the other stations in Ohrid Lake and suppose to be from the mineral dam.

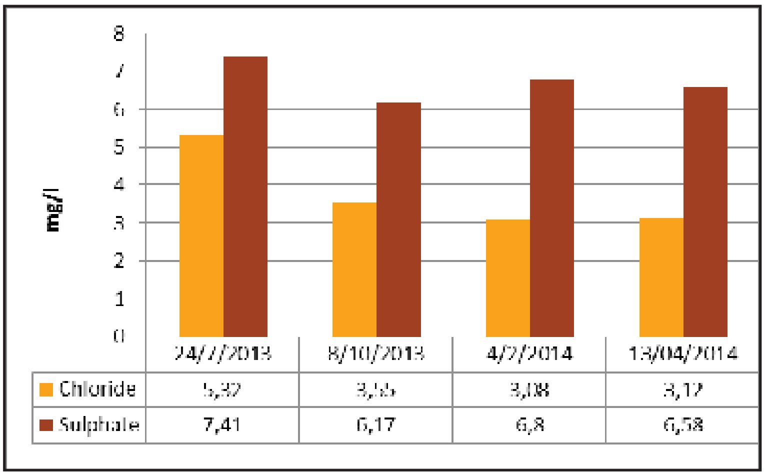


Figure 54. Sulphate and chloride content at Memelishti station water

These parameters are in normal level of the environment of ground of the lake.
The trophic status of the water quality assess by parameters such as transparency (fig. 56), nutrients content (P and N) and the chlorophyll a (fig. 55). All these parameters give an important contribution in assessment of the trophic level of waters. At Memelishti station the values of chlorophyll a are taken in summer and autumn season when the biological activity is more developed. At the figure 55 is presented the values of chlorophyll a parameter in different season and can see the high presence of it in autumn (0.699 mg/l).

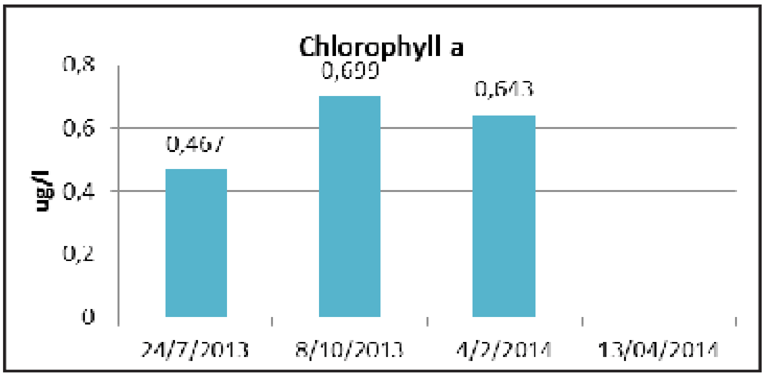


Figure 55. Chlorophyll a content at Memelishti station

Figure 56. Transparency at Memelishti station

Pogradec station

This station is placed on the south part of the lake and the coast of the lake is flat and surrounded by the small agriculture and tourist areas.

The variation of the temperature (fig. 57) is depending from the season and the water flow. It varies from 14.4°C to 25°C. There is a slight difference in conductivity (fig 58) (from 232 to 246 mS/cm) and pH (fig. 59) (from 7.93 to 8.73) that explained the mixing water with Driloni stream that comes into the Ohrid Lake close to this station.

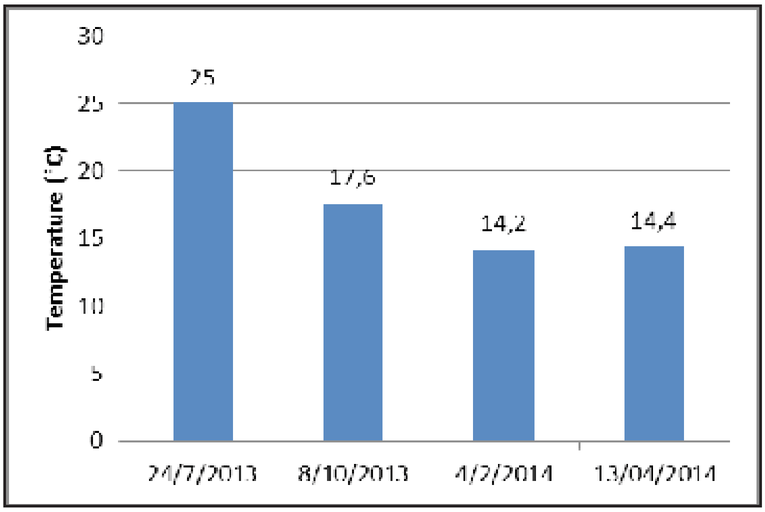


Figure 57. The variation of temperature at Pogradec station

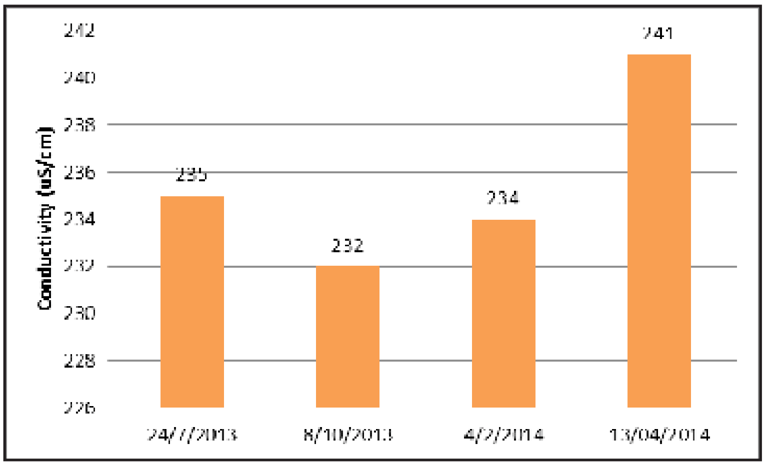


Figure 58. Variation of conductivity at Pogradec station

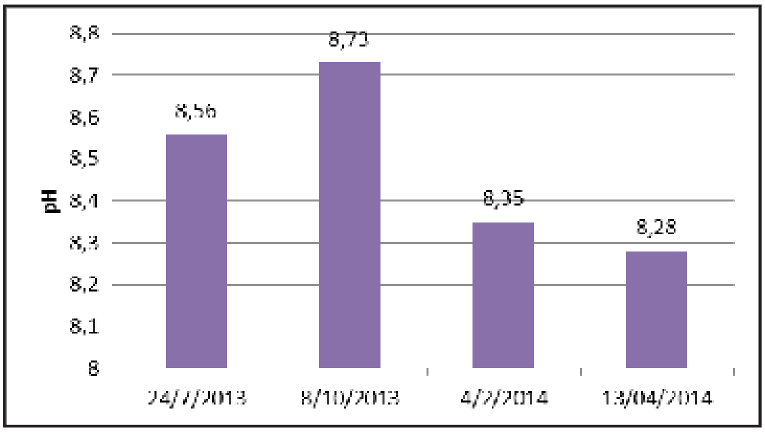


Figure 59. Variation of pH at Pogradeci station

The values of ***pH*** show very well the trend during the different seasons where the difference between summer and winter are small.

The same interpretation we can make even for the ***alkalinity*** of the waters (fig. 60). The values of this parameter vary from 110 mg/l in autumn (October) to 120mg/l in summer (July).

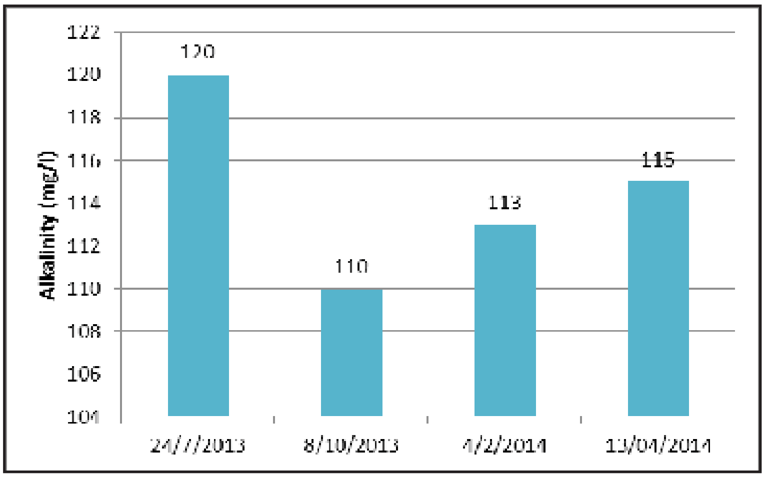


Figure 60. Variation of alkalinity content in water at Pogradec station

Dissolved oxygen shows different values depending on climate indication in water quality. When the temperature is low and the wind is present during the sampling campaign, the results showing the mixing of the waters and small differences of ***DO value from season to season*** (fig. 61).

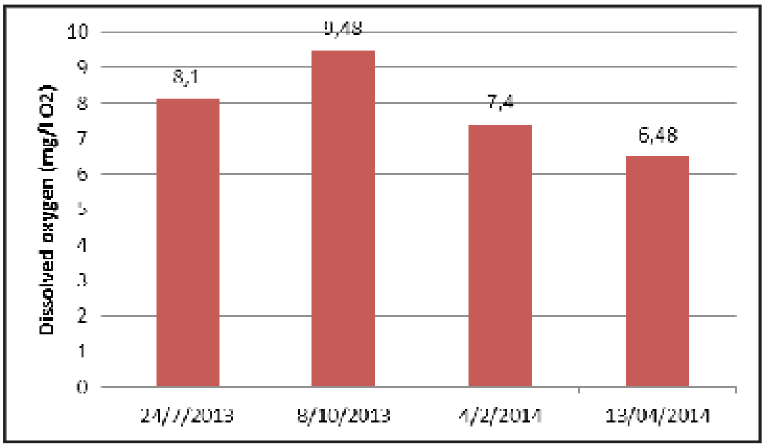


Figure 61. Variation of the dissolved oxygen at Pogradec station

The same results are taken from the ***TC and BOD*** content in waters at Pogradec station (fig.62). The values have variation that depends from the temperature of water and also the biological activities in different seasons.

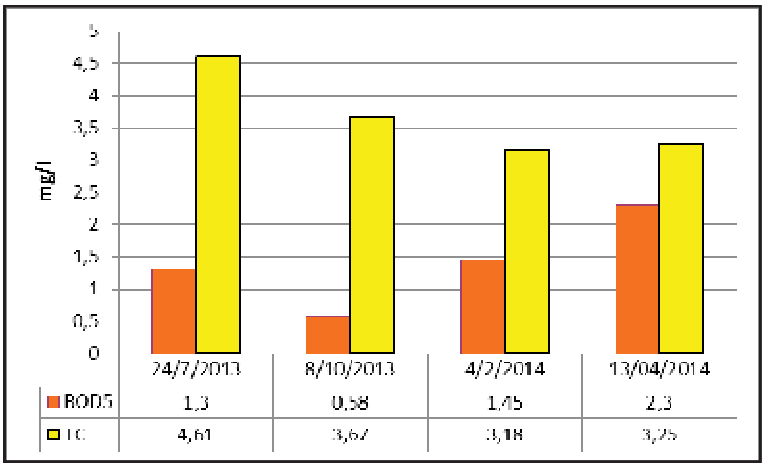


Figure 62. BOD and TC content at Pogradec station

Values of TC show low level of the total carbon present in the water that means low level of organic pollution impact in different seasons. They vary from 3.18 to 4.61 mg/l. Also the values of BOD are lower than the threshold level so the biological impact from the waste water discharges is very low. They vary from 0.58 mg/l O2 in autumn to 2.3 mg/l O2 in spring.

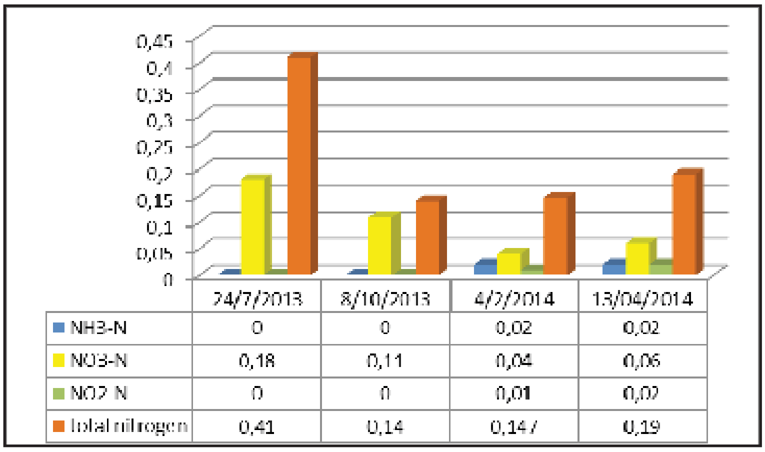


Figure. 63. Nitrogen and its compound content at Pogradec station

Nitrogen content is not high at this station (fig. 63). As we can see the along the year it shows approximately the same values of nitrates content from 0.06 to 0.18 mg N/. The other compounds of nitrogen (ammonia 0.02 mg N/l and nitrites from 0.01 to 0.02 mg N/l) are below the threshold level (< 1).

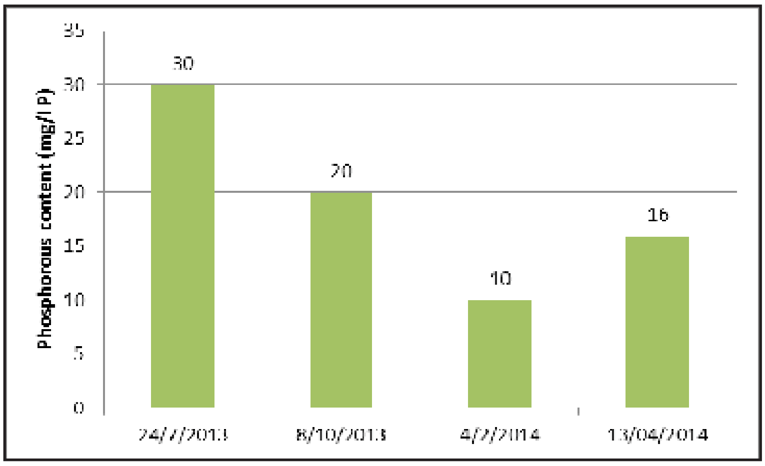


Figure 64. Phosphorous content at Pogradec station

The phosphorous content (fig. 64) is at mesotrophic level. The values (from 10 – 30 mg/l) of ortho phosphorous don't indicate risks in water quality as a limited factor of biological development of water life. Presence of phosphorous in the level from 10 to 20 mg/l P has a negative impact for the good quality of waters.

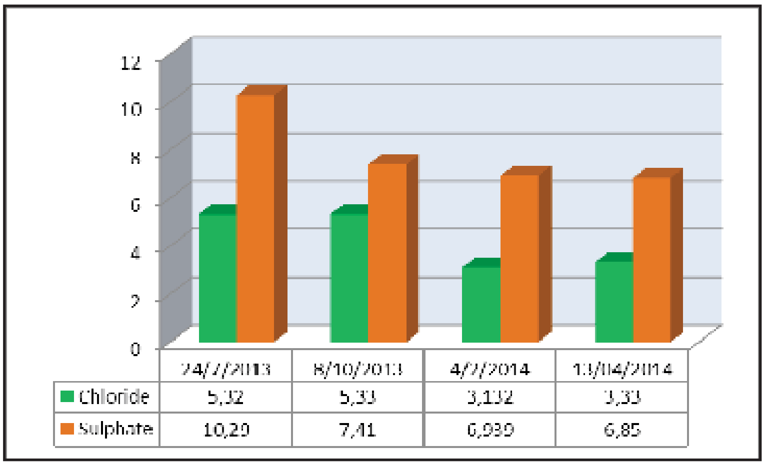


Figure 65. Sulphate and chloride content at Pogradec station water

These parameters play a role in relation of ions balance in water and express the impact from the ground of the lake. Values of these parameters are in normal level (fig. 65).

The trophic status of the water quality assesses by parameters such as transparency (fig. 67), nutrients content (P and N) and the chlorophyll a (fig.66). All these parameters give an important contribution in assessment of the trophic level of waters. At Pogradec station the values of chlorophyll a are taken in summer, autumn and winter season. At the figure 66 is presented the values of this parameter in different season and can see the high presence of it in winter (0.923 mg/l).

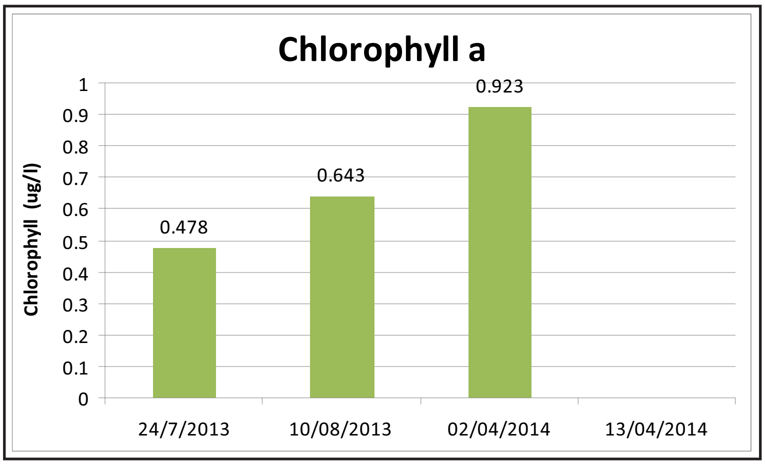


Figure 66. Chlorophyll a content at Pogradec station

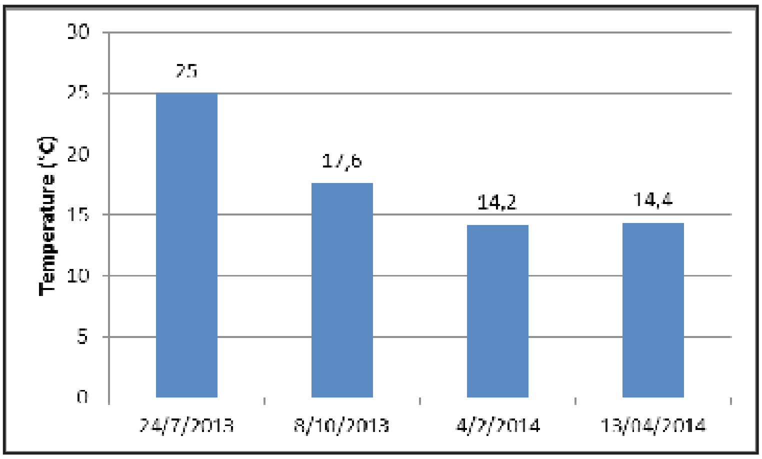


Figure 67. Transparency of water at Pogradec station

The classification of Ohrid Lake water made by using the Carlson Trophic State Index as prescribed below:

Trophic Class	Chlorophyll a content (mg/l)	Phosphorous content (mg/l).	Secchi disc (m)	Trophy Index
Oligotrophic	0—2.6	0—12	>8—4	<30—40
Mesotrophic	2.6—20	12—24	4—2	40—50
Eutrophic	20—56	24—96	2—0.5	50—70
Hypereutrophic	56—155+	96—384+	0.5—<0.25	70—100+

a. TSI for Chlorophyll-a (CA) TSI = 9.81In Chlorophyll-a(ug/L)+30.6

b. TSI for Secchi depth (SD) TSI = 60-14.41In Secchi depth(Meters)

c. TSI for Total phosphorus (TP) TSI = 14.42 In Total phosphorous (ug/l) + 4.15

where TSI is Carlson Trophic State Index and In is Natural logarithm

Carlson's TSI = **[TSI (TP) +TSI (CA) +TSI (SD)] / 3**

where TP and Chlorophyll-a in micrograms/l and SD transparency in meters.

After the calculation of each index

Station	Chlorophyll a content (mg/l)	TSI (CA)	Secchi disc (m)	TSI (SD)	Phosphorous content (mg/l).	TSI (TP)	Trophy Index
Lin	1.91	37	5	37	20	47	40
	0.92	30	4.5	38	10	37	35
	0.594	25			10	37	21
Memelishti	0.467	23	5	37	20	47	36
	0.699	27	5.2	36	20	47	37
	0.643	26	8	30	10	37	31
Pogradec	0.478	23	6	34	30	53	37
	0.643	26	4.3	39	20	47	38
	0.923	30	4	40	10	37	36

Carlson Trophic State Index :

1. *Lin station* - oligotrophic state for TSI
2. *Memelishti station* - oligotrophic state for TSI
3. *Pogradec station* - oligotrophic state for TSI

According the determination of the ecological status of the water bodies physico-chemical elements have only a supporting character.

5.3 Prespa Lake

There are two stations in Prespa Lake involved in monitoring program and for evaluation of the lake water status. The result interpretation will be made station by station and an overall water classification will be discussed for the lake.

Gollomboc stationThis station is placed on the west part of the lake and the coast of the lake is flat and surrounded by agriculture areas that are impacted from the erosion and covered by a lot of plants.

Below we are interpreted the results taken from the analyses of the water samples taken in different seasons according to the monitoring program implementation.

The variation of the temperature is depending from the season and varies from 11.1°C to 27°C. Prespa Lake is a shallow lake and the water circulates during the year. Also other water quality parameters such as pH (fig. 68) (from 8.06 to 8.77) and conductivity (fig. 69) (from 216 ms/cm to 277 ms/cm) have small differences of values during the year.

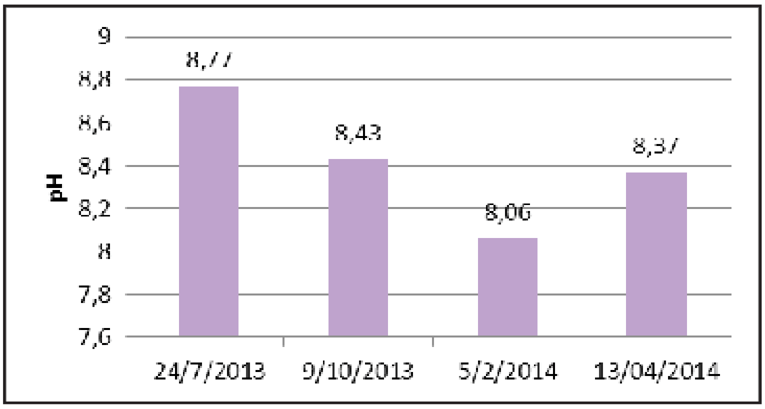


Figure 68. Variation of pH at Gollomboc station

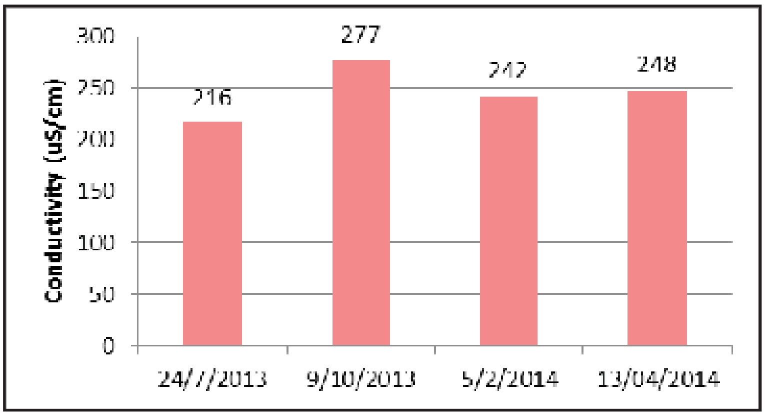


Figure 69. Variation of conductivity at Gollomboc station

The ***pH*** variation shows very well the changes between the climate conditions during the year and they are depended by season temperature.

The same discussion we cannot make for the ***alkalinity*** of the waters (fig. 70). The values of this parameter have differences in different season. It varies from 98 mg/l in autumn (October) to 113mg/l in spring (April).

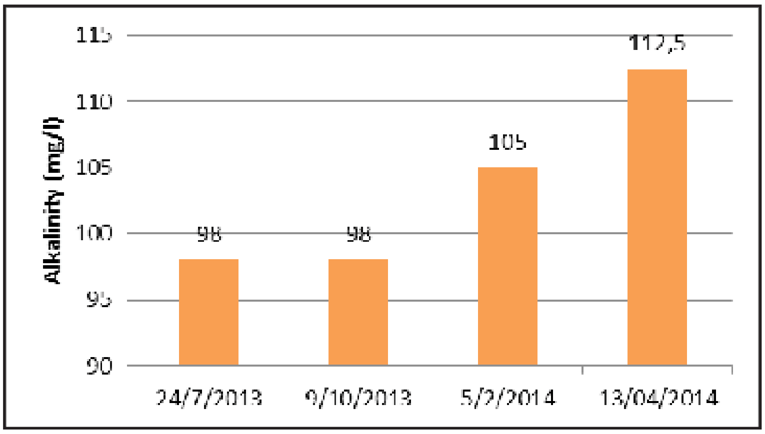


Figure 70. Variation of alkalinity content in water at Gollomboc station

Water of lake is well oxygenated in different season of the year even there are some climate indications in water quality (sources from NEA monitoring data). But the data collected in campaign shows a non correct measurement of this parameter in site. According to the results we can see that in different seasons the DO content has big differences (7.5 mg/l O2 in July and 14.63 mg/l O2 in April).

The results taken for ***TC and BOD*** content in waters at Gollomboc station (fig.71) presented the same situation in different season. The values of TC have variation from 4.64 mg/l in summer to 6.04 mg/l in autumn that depends from the temperature of water and also the biological activities in different seasons. The values of BOD vary from 0.42 mg/l O_2 in autumn to 2.53 mg/l O_2 in spring that shows the biological activities during these seasons.

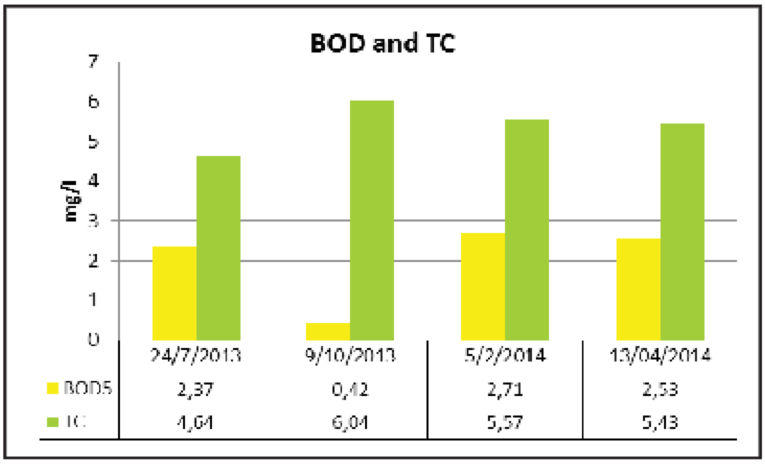


Figure 71. COD, BOD and TC content at Gollomboc station

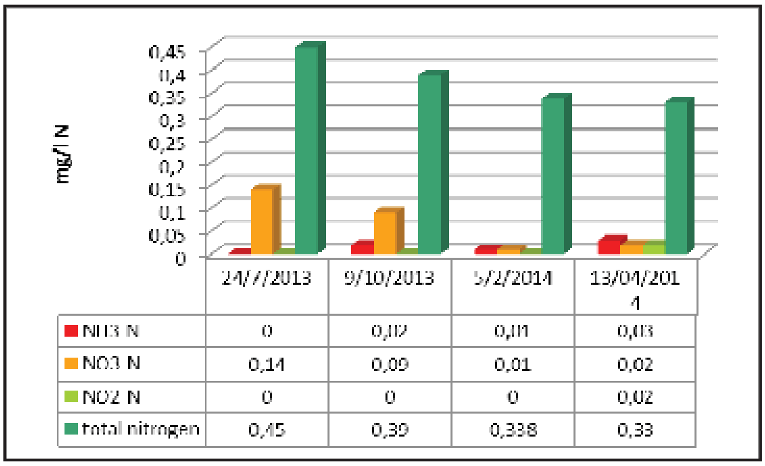


Figure 72. Nitrogen and its compound content at Gollomboc station

Nitrogen content is not high at this station (fig. 72). The values of nitrates content vary from 0.01 to 0.14 mg N/l at the winter and summer season. The other compounds of nitrogen like ammonia vary from 0.01 to 0.03 mg N/l and they are below the threshold level (< 1).

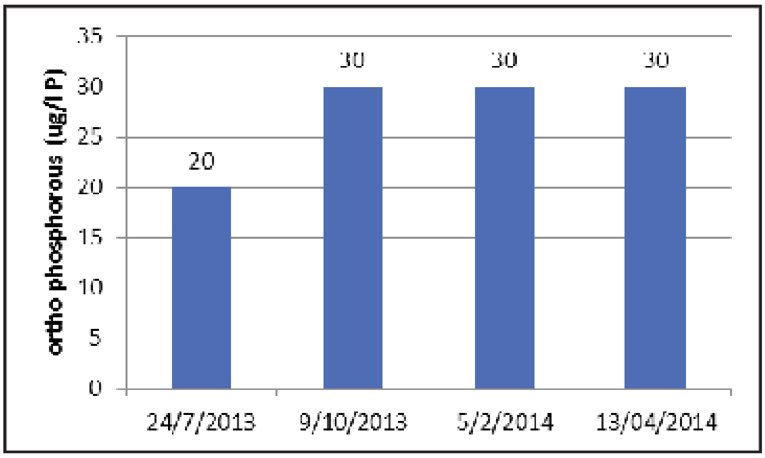


Figure 73. Phosphorous content at Gollomboc station

The phosphorous content (fig. 73) is at mesotrophic level. The values (from 20 – 30 mg/l P) of ortho phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Presence of phosphorous in this level has a negative impact for the good quality of shallow waters that are impacted from urban discharges and agriculture fertilizer.

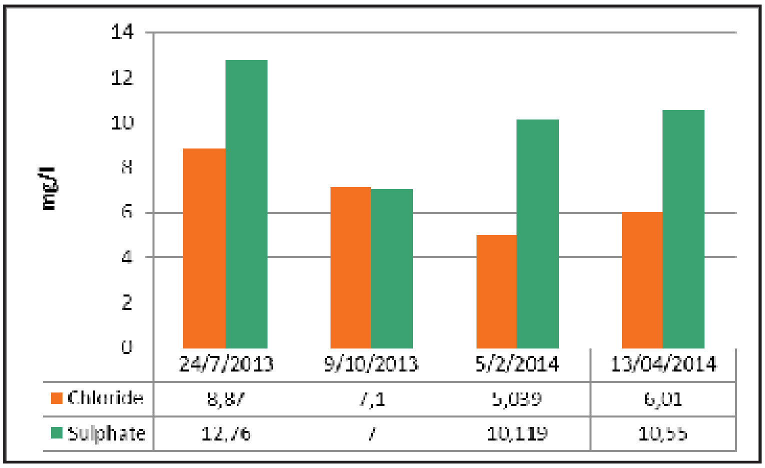


Figure 74. Sulphate and chloride content at Gollomboc station

These parameters are significant for the assessment of the water ions balance and express the impact from the ground of the lake. Values of these parameters are in normal level for the environment of ground of Prespa Lake.

The trophic status of the water quality assess by parameters such as transparency (fig. 76), nutrients content (P and N) and the chlorophyll a (fig. 75). All these parameters give an important contribution in assessment of the trophic level of waters. At Gollomboc station the values of chlorophyll a are taken in different seasons. At the figure 75 is presented the values of this parameter in different season and can see the high presence of it in autumn (1.412 – 5.116 mg/l).

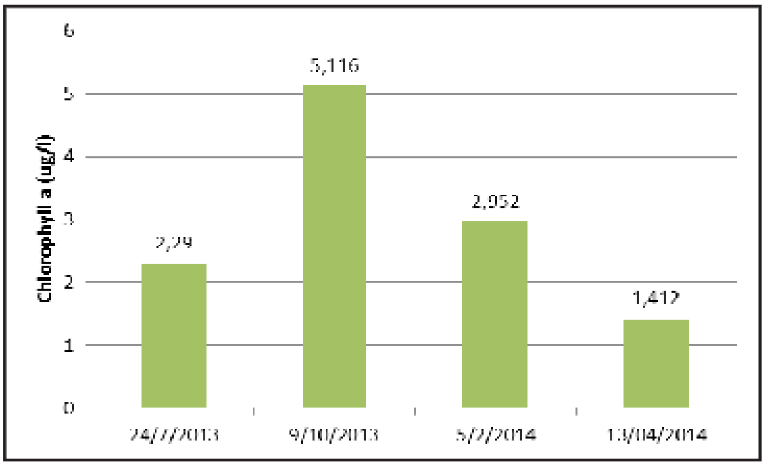


Figure 75. Chlorophyll a content at Gollomboc station

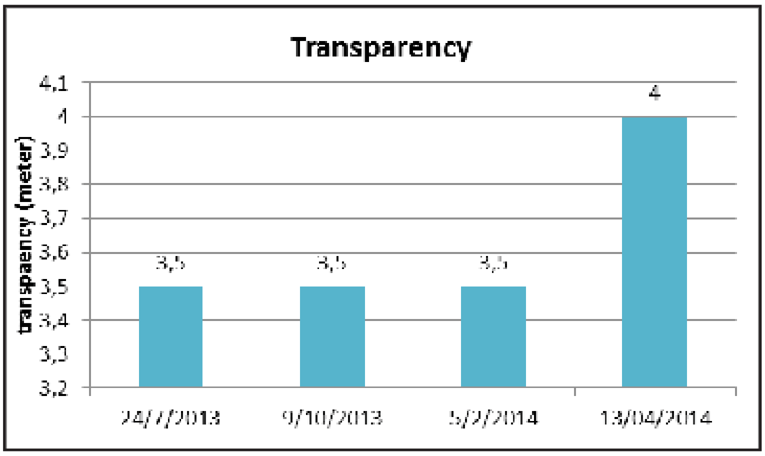


Figure 76. Transparency of the water at Gollomboc station

Pustec (Liqenas) station

This station is placed on the west part of the lake and the coast of the lake is gravel and rocky. This part of the lake is impacted by the mineral damp and the mine stream flows.

The variation of the temperature is depending from the season. The layers of water column have small differences for water quality parameters such as pH (fig 77) (from 7.8 to 8.62) and conductivity (fig. 78) (from 209 ms/cm to 248 ms/cm).

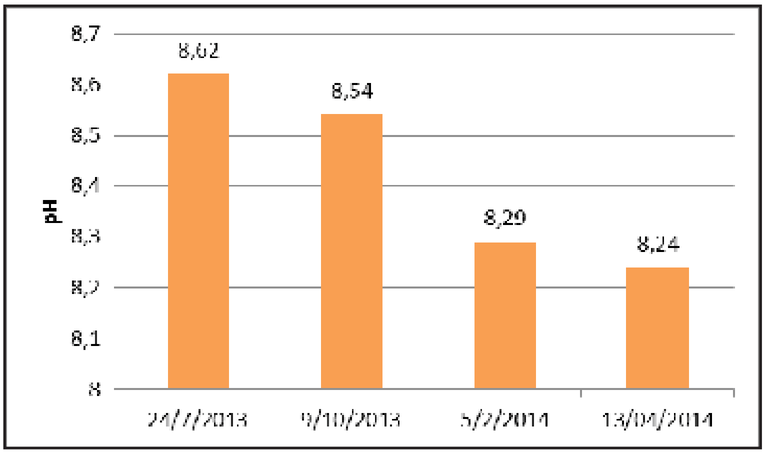


Figure 77. Variation of pH at Pustec station

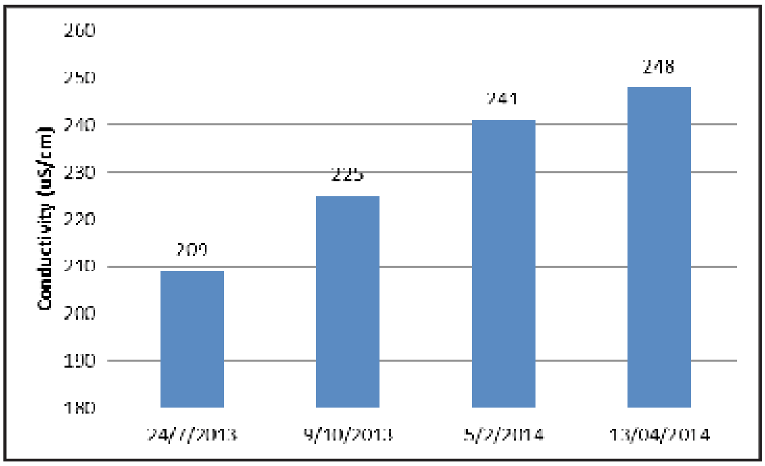


Figure 78. Variation of conductivity at Pustec station

The ***alkalinity*** of the waters (fig.79) has no significant differences in different season. It varies from 93 mg/l in summer (July) to 112.5 mg/l in spring (April).

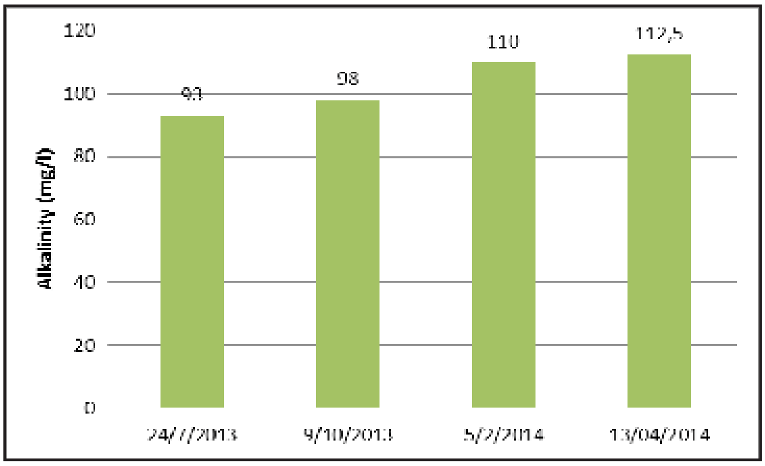


Figure 79. Variation of alkalinity content in water at Pustec station

Results of DO content show they depended from the climate indication in water quality in different season of the year. The data collected in campaign shows a non correct measurement of this parameter in site. According to the results we can see that in different seasons the DO content has big differences

(7.71 mg/l O2 in July and 11.26 mg/l O2 in April).

The results of the ***TC and BOD*** content in waters at Pustec station are shown in figure 80. The values have variation that depends from the temperature of water and also the biological activities in different seasons. For the TC values vary from 5.55 (in winter) to 5.95 mg/l (in autumn) and for BOD vary from 0.03 (in winter) to 5.6 mg/l O_2 (in spring).

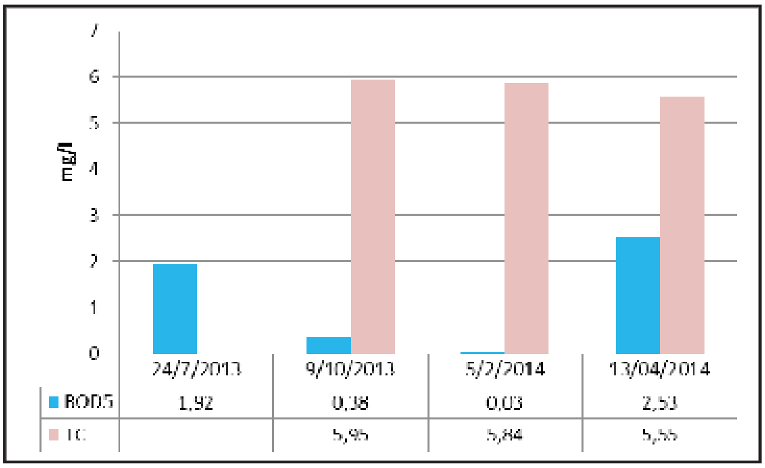


Figure 80. BOD and TC content at Pusteci station

The values of BOD are at second threshold level (3 -5.5 mg/l O_2) so the biological impact from the waste water discharges is indicated at mesotrophic level.

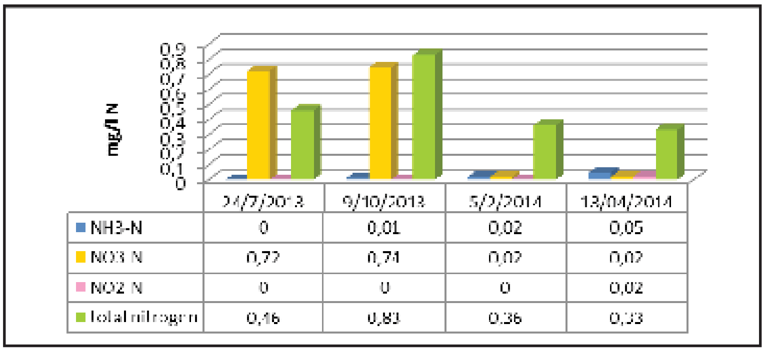


Figure 81. Nitrogen and its compound content at Pusteci station

Nitrogen content is at the same level as other station. As we can see the values of nitrates content vary from 0.02 (spring) to 0.74 mg N/l (in autumn). The other compounds of nitrogen ammonia and nitrites are at the lowest level. They are below the threshold level (< 1).

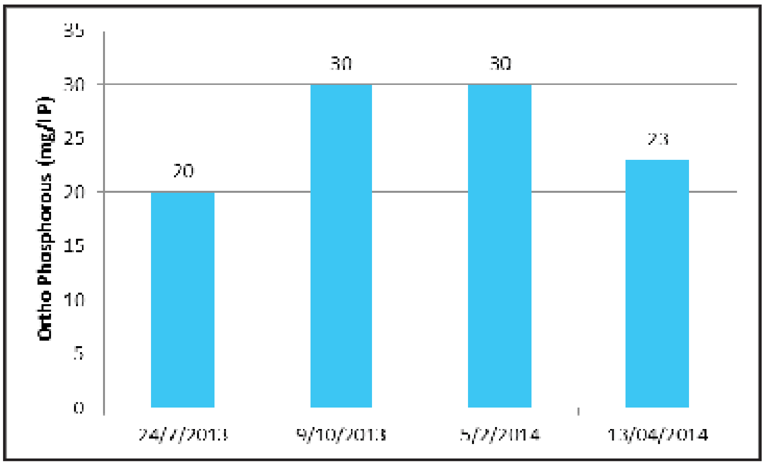


Figure 82. Phosphorous content at Pusteci station

The phosphorous content (fig. 82) is at mesotrophic level. High values (from 20 – 30 mg/l) of ortho phosphorous indicate a risk in water quality as a limited factor of biological development of water life. Presence of phosphorous in this level is the same as the other station in Prespa Lake and supposes to be from agricultural areas.

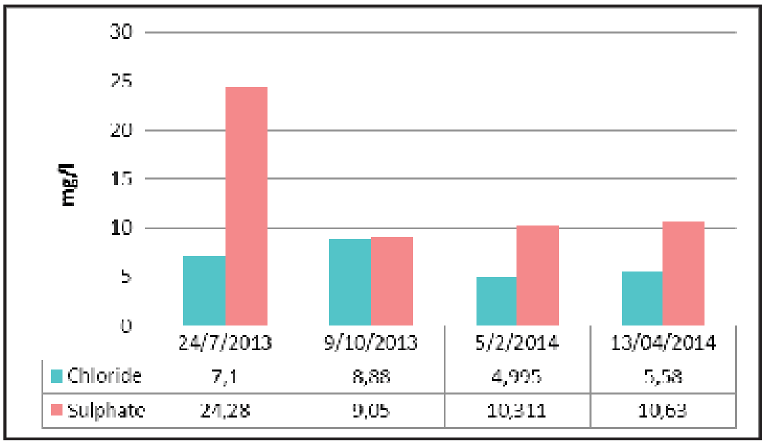


Figure 83. Sulphate and chloride content at Pusteci station water

These parameters are in normal level of the environment of ground of the lake.
The trophic status of the water quality assess by parameters such as transparency (fig. 85), nutrients content (P and N) and the chlorophyll a (fig. 84). All these parameters give an important contribution in assessment of the trophic level of waters. At Pusteci station the values of chlorophyll a are taken in different seasons. At the fig. 84 is presented the values of chlorophyll a parameter in different season and can see the high presence of it in autumn (5.811 mg/l).

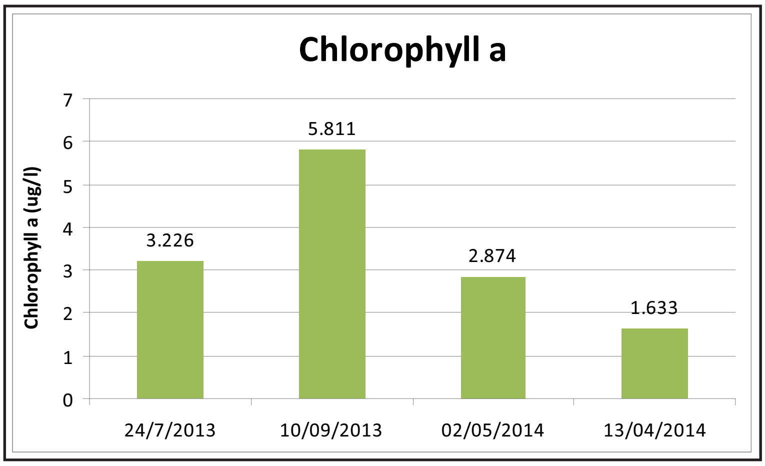


Figure 84. Chlorophyll a content at Pusteci station

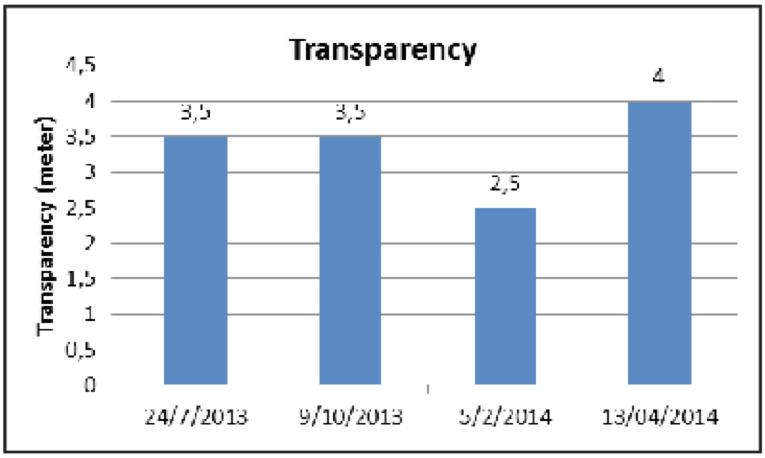


Figure 85. Transparency at Pusteci station

The classification of Prespa Lake water made by using the Carlson Trophic State Index as prescribed below:

Trophic Class	Chlorophyll a content (mg/l)	Phosphorous content (mg/l).	Secchi disc (m)	Trophy Index
Oligotrophic	0—2.6	0—12	>8—4	<30—40
Mesotrophic	2.6—20	12—24	4—2	40—50
Eutrophic	20—56	24—96	2—0.5	50—70
Hypereutrophic	56—155+	96—384+	0.5—<0.25	70—100+

a. TSI for Chlorophyll-a (CA) TSI = 9.81In Chlorophyll-a(ug/L)+30.6

b. TSI for Secchi depth (SD) TSI = 60-14.41In Secchi depth(Meters)

c. TSI for Total phosphorus (TP) TSI = 14.42 In Total phosphorous (ug/l) + 4.15

where TSI is Carlson Trophic State Index and In is Natural logarithm

Carlson's TSI = [TSI (TP) +TSI (CA) +TSI (SD)] / 3

where TP and Chlorophyll-a in micrograms/l and SD transparency in meters.

After the calculation of each index (table below):

Station	Chlorophyll content (mg/l)	TSI (CA)	Secchi disc (m)	TSI (SD)	Phosphorous content (mg/l).	TSI (TP)	Trophy Index
Gollomboc	2.290	39	3.5	42	20	47	43
	5.116	47			30	53	33
	2.952	41	3.5	42	30	53	45
	1.412	34	4	40	30	53	42
Pustec (Liqenas)	3.226	42	3.5	42	20	47	44
	5.811	48			30	53	34
	2.874	41	2.5	47	30	53	47
	1.633	35	4	40	23	49	42

Carlson Trophic State Index :

Gollomboc station - mesotrophic state for TSI

Pusteci (Liqenas) station - mesotrophic state for TSI

According the determination of the ecological status of the water bodies physico-chemical elements have only a supporting character.

6. Recommendations

This project has been a good collaboration between all experts, organizations and NEA for the collaboration between national and international organization involved in this process. This experience should be continued during the following Monitoring Programme according the requirements of the WFD under the leadership of the NEA.

Therefore it is necessary to do transboundary agreements concerning the location of monitoring stations, parameters, frequency, methods of sampling and analyzing, assessment procedures considering the monitoring of the biological elements.

The implementation of the new law for Integrated Management of Water Resources needs to identify the human capacities and to coordinate the work between different monitoring organization that are or not responsible for implementation of the environment monitoring issues (physico-chemical, biological, microbiological and other important parameters as are prescribed in WFD requirements).

Report
of
physical-chemical and microbiological investigations of Skadar Lake water and sediment quality
(Montenegrin side)

Prepared by: MSci Pavle Djuraskovic

Podgorica, June 2014

Contents

1. Introduction

Realization of the project ``Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar (herewith to CSBL)- Component 4: Monitoring of water quality of Skadar Lake pursuant of EU Water Framework Directive (WFD), was based on the MoU, signed on 6th November 2012, and Contract 83129311, signed on 21st November 2012, between Deutsche Gesellschaft für Internationale Zusammenarbeit GmBH (hereinafter GIZ) and Institute of Hydrometeorology and Seismology (hereinafter IHMS).

The main goal of the cooperation is to assess the current ecological and chemical status of the water bodies of Skadar Lake as one of several requirements under the WFD.

Specific objectives were as follow:

- strengthen the capacity of IHMS to conduct baseline analyses for lake characterization and to implement surveillance, operational and/or investigative monitoring;
- implement jointly *Component 4* – Lake Characterization and Water Framework Directive – of the Plan of Operations (PoO) of the CSBL Project;
- Improve transboundary collaboration.

The cooperation represents one of several technical assistance components outlined in the Agreement. The scope of the cooperation in terms of parameters to be monitored under the current MoU is defined in consultation with CSBL and the Technical Working Group WFD which was established on September 19, 2012 and of which IHMS is a member.

2. Selection of the sampling points

The sampling points for water quality measuring were selected in accordance with project tasks, WFD requirements and IHMS experience in previous regular monitoring. Also it is taken into consideration the sampling network that should be selected on the Albanian side.

Six sampling points were selected in accordance with project purposes: Kamenik, Virpazar, Plavnica, Podhum, Starcevo and Center (see Table 1.). They cover the target parts of the Lake. Both, littoral and pelagic zone of the Lake were covered by the selected points. The sampling points at littoral have corresponded with expected anthropogenic impact sites and sources.

The farthest point of Montenegrin side, in the same time the border point, Ckla was omitted because so short distance from the neighbouring Albanian point Zogaje. However, the site has to be included in the future monitoring program, regarding Article 8 WFD, as a cross-border point. The network should cover the water bodies of the Lake. The measuring data should be used for initial characterisation of lakes` water bodies. The characterisation of selected stations is presented by each point, as follow.

KAMENIK. It is situated at north-west part of the Lake, a closed water area, named as Vucko blato. This area is under the biggest anthropogenic pressure. Here are mixed polluted waters of Moraca-right branch and the river Crnojevica with pure waters of Bisevina and Karatuna rivers and numerous sub-lacustrine springs (Karuc, Djurovo oko, Bazagur, Ploca, Grab, Poseljane etc.). This is area of possible inflow of leaching waters from Cetinje dump. The site is near the island Kamenik, at the water stream and boat waterway. There are rich colonies of floating vegetation at the surrounding. The depth of water is moderate.

VIRPAZAR. It is situated in front of mouth of the river Orahovstica (i.e. Virpazar channel), on the waterway, surrounded with large colonies of floating vegetation. The site belongs to littoral zone. The water depth at site is low.

PLAVNICA. The point is in front of mouth of the river Plavnica (channel). The wider water area presents a typical littoral zone, with the shallowest water in the Lake. The impact of polluted ground water (inorganic and organic toxic pollutants) is expected at this location. The added contribution can be from frequent traffic of many small motor fishing boats and the restaurant, nearby. The water surrounding is covered by floating vegetation. Wide open coast can fully communicate with pelagic area.

PODHUM. It is located at closed estuary, down the Hum hill. The water depth is moderate, but the surrounded coast is low, overgrown with lake vegetation. The main activity at the wider land area is agriculture. The water surrounding is covered by large colonies of floating vegetation. There are no local pollution sources, close to this site, but the possible pollution influence by underground waters is expected.

STARCEVO. It is located near to island Starcevo, between the island and the coast, at the main stream from Moraca to Bojana. There are no local pollution sources nearby. The transport of pollution from upstream area can be monitored at this station. The water is relatively deep, without floating vegetation around.

Table 1.: Selected measuring points

M. point	ID	GIS	Water area	Impact
Kamenik	MNE1	N 42° 17´ 32.63˝ E 19° 06´ 3.10˝	Coastal	Tributaries: Crnojevica r., Moraca-right stream; possible ground waters from Cetinje dump
Virpazar	MNE2	*N 42° 15´ 22.12˝* *E 19° 06´ 19.85˝*	Littoral	Settlement Virpazar, by Orahovstica r.
Plavnica	MNE3	N 42° 15´ 40.26˝ E 19° 11´ 54.02˝	Littoral	Ground waters from Zeta plane: agriculture and industry
Podhum	MNE4	N 42° 16´ 2.58˝ E 19° 21´ 44.53˝	Coastal	Possible impact of ground waters from Zeta plane: agriculture and industry
Starcevo	MNE6	N 42° 11´ 19.82˝ E 19° 12´ 29.42˝	Coastal	Transport of pollution during extended stream of Moraca
Center	MNE1	N 42° 10´ 47.50˝ E 19° 16´ 26.67˝	Pelagic	State of water quality of main water body

CENTER. This point belongs to pelagic zone, located near of the border corner at the lake. The site represents a resulting state of the water quality, characteristic for main water mass of the lake. This area has the deepest water.

Picture 1.: Measuring sites of CSBL at Montenegrin side

3. Description of the sampling campaigns (weather conditions etc)

Dates of sampling were chosen when meteo-hydrological and ecological conditions corresponded to characteristics of the related season. However, the hydrological year was not typical, especially in winter campaign, when temperature of water and air were higher, but water level was lower, as usually.

Sampling of water quality was conceived on seasonal base. Sampling activity took two days for each campaign.

The first campaign, in spring, dated on 17-18 April 2013; the second, in summer, dated on 18-19 July 2013; the third, in autumn, dated on 22-23 October 2013; the forth, in winter, dated on 7-8 February 2014. Sampling of sediment was done during summer season. The inter-comparison exercise with Albanian partners, due to quality assurance, provide by the same (summer) period.

Organoleptic characteristics of water were satisfied: color of water was in natural, green shades; specific smell and taste were not perceptible. Floating materials were not observed, except remains of water flora.

Spring campaign occurred after relatively long rainy period. The weather was sunny, with moderate air temperature and without wind (calma). Air temperature was increasing from 17,8°C to 29,8°C, depending of sampling time and the day of field work. The perceptible water color was typical at each measuring area. The water was odorless and tasteless. The residue of vegetation nor other floating waste was not observed.

The summer campaign was realized in warm weather, with air temperature 27,5 °C to 32,0°C. The weather was sunny, with moderate air temperature and without wind, and consequently, without water waves.

The autumn campaign was done in sunny weather, with moderate air temperature (between 17-20°C) and without wind. Air temperature was very similar with water temperature.

The winter campaign was realized in stabile weather. Air temperature was about 10°C (8,2-10,7°C), like water temperature. The overall weather in longer previous period has corresponded to extended autumn conditions rather than winter.

4. Methods for sampling, transport and storage, analysis

The sampling of water at all measuring stations was carried out in four campaigns, which are belonged to four different seasons, what is recommended of WFD: Aril, July, October and February. Collecting of samples was done by motor-bout, owned of National park ``Skadar Lake``. Two days were spent for collection of samples in each campaign. Samples of water were taken by Ruthner bottle of 5lit, which was provide by the Project. Water samples were taken in two depths down the water column (0,5m below surface and 1m above bottom). All sampling procedures conducted according official standard methodology. Samples for general chemical analysis were collected in containers of inert plastic, with 3lit volume. Samples for oxygen and BOD_5 were collected in Winkler bottles of 100ml. The samples for oxygen and BOD_5 were preserved with $MnCl_2$ and KJ. Samples for chlorophyll-a are collected in dark glass bottles of 1lit. Some volume of water was filtered through the glass filter 0,45µm. Samples for microbiology were collected in sterile dark glass bottles of 250ml.

Some parameters were measured in situ, as follow: Temperature of water and air, depth, transparency, pH, conductivity, as well as observation of hydro-meteo conditions and organoleptic state.

Sampling of sediment was done during July campaign, in the same time as water sampling, only one per overall measuring period, because stability of the content of these specific pollutants during the time. All sampling sites, have been affected by the impact of the pollution by Bojana outflow. According to the WFD requirements analysis of the sediment considered the main pollutants of the discharges to the lake, especially heavy metals (Hg, Cd, Pb) and organo-chloride pesticides. The sediment samples were taken by a Van Veen grab. About 1/2kg of captured raw sediment materials was collected into plastic boxes after removal of vegetation residues. The samples were not prepared with any reagents.

Bottles with samples were disposed in hand refrigerators, at low temperature. The samples were transported to laboratory by car. The containers with samples were saved of any outside influences. The samples were transported to laboratory as soon as possible. After their arrival the samples were disposed in refrigerators at 4°C, before analysis.

All analyses were done by standard methods. The measuring was done in situ and in laboratory. Temperature of water and air, depth, transparency, pH and conductivity were measured on site.

All applied analytical methods and other procedures have been harmonized with related standards (ISO, EPA and other).

The sediment samples for heavy metals content were analysed by ICP-OES method. The pesticides were measured by GC.

Table 2.: Analytical methods and techniques used

Parameters		Method	Technique
Turbidity		APHA-AWA-WPCF, 1975	Turbidimeter
Temperature		APHA-AWA-WPCF, 1975	Termometer
Transparency		APHA-AWA-WPCF, 1975	Secchi disk ø 20cm
Conductivity		(APHA-AWA-WPCF, 1975)	Conductometry
pH		(APHA-AWA-WPCF, 1975)	Ion selective glass electrode
Alkalinity		(APHA-AWA-WPCF, 1975)	Titrimetry
TSS		(APHA-AWA-WPCF, 1975)	Gravimetry, drying at 105°C
DO		APHA-AWA-WPCF, 1975	Acc. Winckler method
Saturation of O2			Calculation
COD		``Water for drink`` JUS standards 1991	Dissolve by permanganate consumption
BOD5		APHA-AWA-WPCF, 1975	Acc. Winckler method
Ammonia-Nitrogen		APHA AWWA WPCF, 1975	VIS spectrophotometry, 630nm
Nitrates-Nitrogen		APHA-AWA-WPCF, 1975	UV spectrophotometry, 220nm
Nitrites-Nitrogen		APHA-AWA-WPCF, 1975 meth. No 420	VIS spectrophotometry, 520nm
TN		EPA 415	Thermo-cathalytic oxidation
Orto-Phosphorus		APHA AWWA WPCF 1975	VIS spectrophotometry, 690nm
Total Phosphorus		APHA AWWA WPCF 1975	VIS spectrophotometry, with $K_2S_2O_8$ at 690nm
TOC		EPA 415	Thermo-cathalytic oxidation
Chlorides		APHA-AWA-WPCF, 1975	Volumetry
Sulfates		APHA AWWA WPCF, 1975	UV spectrophotometry, 400nm
Fluorides		ISO 10304-1	Ion selective electrode
Chlorofill-a		ISO 10260 (1992)	VIS spectrophotometry
Mercury		EPA 7473	DMA
Arsenic		MW digestion	HGS
Lead		ISO 11466.3	ICP-OES
Cadmium		ISO 11466.3	ICP-OES
Alumina		ISO 11466.3	ICP-OES
Pesticides	Organo-chlorine	EPA 3535 EPA8081B	GC/ECD
	Organo-phosphorus	EPA 3535 EPA8141B	GC/NPD
Coliform Bacteria		JUS standard in ``Water to drink`` Ministry of Health SRJ	Membrane filtration
Faecal Bacteria		ISO 9308-1	Membrane filtration
Heterotrophic Bacteria		ISO 6222	Membrane filtration

5.1. Results (tables and graphics)

Water quality

All gathered results for water and sediment quality presented in the next tables and graphics.

Table 3.1.: Measured parameters and their detection limits

Parameters	Abbreviation	Medium	m. unit	Detection limit - DL
Depth	D		m	0.1
Turbidity	T	Water	ntu	0.20
Temperature	Temp	Water	°C	0.1
Transparency by Seeki	Trans	Water	m	0.1
Conductivity	Cond	Water	µS/cm	1
pH	pH	Water	-	0.01
Alkalinity	Alk	Water	ml HCl	0,1
Total Suspended Solids at 105°C	TSS,	Water	mg/l	0,1
Dissolved Oxygen	DO	Water	mgO2/l	0,01
Saturation of O2	SAT	Water	%	1
Chemical Oxygen Demand	COD	Water	mgO2/l	0.01
5-days Biochemical Oxygen Demand	BOD_5	Water	mgO2/l	0.01
Ammonia-Nitrogen	NH_3-N	Water	mgN/l	0,01
Nitrates-Nitrogen	NO_3-N	Water	mgN/l	0.001
Nitrites-Nitrogen	NO_2-N	Water	mgN/l	0.001
Total Nitrogen	TN	Water	mgN/l	0.001
Orto-Phosphorus	o-PO_4-P	Water	µgP/l	1
Total Phosphorus	TP	Water	µgP/l	1
Total Organic Carbon	TOC	Water	mg/l	0,01
Chlorides	Cl	Water	mg/l	0.1
Sulfates	SO_4	Water	mg/l	0.1
Fluorides	F	Water	mg/l	0,001
Chlorofill-a	Chl-a	Water	mg/l	0,01
Mercury	Hg	Sediment	µg/kg	0.01
Arsenic	As	Sediment	mg/kg	0.001
Lead	Pb	Sediment	mg/kg	0.01
Cadmium	Cd	Sediment	mg/kg	0.01
Alumina	Al	Sediment	g/kg	0.01
Pesticides	Pest	Sediment	µg/kg	0.0125
Total number of Coliform Bacteria	TC	Water	nfu/100ml	-
Total number of Faecal Bacteria	TFC	Water	nfu/100ml	-
Total number of Heterotrophic Bacteria	HA	Water	nfu/1ml	-

Table 3.1.1: Water quality results for Kamenik station – MNE 1

KAMENIK								
Water layer Parameters	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom
date of sampling	18.04. 2013	18.04. 2013	19.07. 2013	19.07. 2013	23.10. 2013.	23.10. 2013.	7.02. 2014.	7.02. 2014.
Depth, m	-	6.6	-	4,3	-	6.0	-	6.0
Sampling depth, m	0,5	5.5	0,5	3,5	0,5	5	0,5	5
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	2,68	< DL	< DL
Temperature, °C	16.0	13.0	20,4	20,5	15,8	12.0	9,6	9,4
Transparency, m	3,8	-	2,4	-	3,4	-	3,3	-
Conductivity, µS/cm	222	224	252	244	261	260	235	249
pH	8,14	8,17	7,98	8,01	7,65	7,78	7,84	7,73
Alkalinity, ml HCl	3,0	2,7	2,6	2,6	3,1	-	2,7	2,9
TSS, mg/l	< DL	< DL	0,2	45,6	0,2	2,6	1.0	0,1
DO, mgO2/l	10,96	12,29	9,10	10.00	9,11	9,62	11,8	11,2
O_2 Saturation, %	112	117	109	112	92	89	104	98
COD, mgO2/l	1,37	1,19	2,90	1,60	2,27	1,74	1,20	1,40
BOD_5, mgO2/l	1,95	3,80	2,10	2.00	2,42	2,14	2.00	1,50
NH_3-N, mg/l	0,06	0,04	0,05	0,01	< DL	0,03	0,055	0,023
NO_3-N, mg/l	0,108	0,336	0,018	0,014	0,240	0,570	0,133	0,143
NO_2-N, mg/l	0,003	0,002	< DL	0,001	0,007	0,008	0,003	0,001
TN, mg/l	0.492	0.775	-	-	0,510	0,710	0,250	0,180
o-PO_4-P, µg/l	4	10	10	3	12	16	16	< DL
TP, µg/l	18	11	43	30	16	19	17	10
TOC , mg/l	1.28	1.23	-	-	2,32	1,19	1,45	1,06
Cl, mg/l	2,1	2,1	4,2	3,9	3,4	1,9	4,1	2,3
SO_4, mg/l	4,4	4,9	4,5	5.0	6.0	6.0	3,5	4,4
F, mg/l	0.620	0.063	-	-	1,780	0,074	0,053	0,053
Chl-a, mg/l	6,20	-	8,05	-	1,79	-	10,08	-
TC, nfu/100ml	19	86	8	79	6	535	3	2
TFC, nfu/100ml	10	64	9	20	2	425	0	0
HA, nfu/1ml	7	9	3	19	12	123	10	4

Table 3.1.2.: Water quality results for Virpazar station – MNE 2

VIRPAZAR								
Water layer Parameters	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom
date of sampling	18.04. 2013	18.04. 2013	19.07. 2013	19.07. 2013	23.10. 2013.	23.10. 2013.	7.02. 2014.	7.02. 2014.
Depth, m	-	5,75	-	3.0	-	3,8	-	5.0
Sampling depth, m	0,5	4,75	0,5	2.0	0,5	2,8	0,5	4.0
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
Temperature, °T	15.0	13.0	26,1	24,3	13,8	12,4	9,6	9.0
Transparency, m	2,8	-	1,9	-	2,6	-	1,8	-
Conductivity, µS/cm	231	226	225	248	266	263	270	273
pH	8,11	8,07	8,15	7,95	7,87	7,92	7,88	7,87
Alkalinity, ml HCl	2,8	2,8	2,7	2,3	3,2	-	2,8	3,1
TSS, mg/l	< DL	< DL	0,3	10	0,2	1,2	0,6	2,1
DO, mgO2/l	12,08	11,75	10,40	9,80	9,55	10,15	12,20	11,90
O_2 Saturation, %	120	112	125	118	93	95	107	103
COD, mgO2/l	1,35	1,18	1,90	2,50	2,70	1,94	2,70	1,94
BOD_5, mgO2/l	3,28	2,64	2,90	1,40	3,59	1,98	2,30	0,70
NH_3-N, mg/l	0,12	0,15	0,05	0,06	0,05	0,03	0,07	0,03
NO_3-N, mg/l	0,232	0,481	< DL	< DL	0,320	0,580	0,165	0,185
NO_2-N, mg/l	0,002	0,003	< DL	0,001	0,006	0,009	0,003	0,002
TN, mg/l	0.995	0.828	-	-	0,500	0,690	0,300	0,240
o-PO_4-P, µg/l	10	16	13	20	15	16	13	13
TP, µg/l	22	24	13	30	17	19	0,023	0,033
TOC , mg/l	2.07	1.64	-	-	1,88	1,31	1,52	1,18
Cl, mg/l	3,1	3.0	3,5	4.0	3,1	1,6	3,5	3,9
SO_4, mg/l	4,5	4,8	4,5	4,1	5,5	5,4	4,7	5,9
F, mg/l	0.062	0.066	-	-	1,680	1,860	0,044	0,043
Chl-a, mg/l	5,33	-	5,63	-	0,20	-	7,02	-
TC, nfu/100ml	63	170	57	145	36	450	13	36
TFC, nfu/100ml	27	150	27	59	34	410	2	7
HA, nfu/1ml	7	12	53	29	5	15	4	12

Table 3.1.3.: Water quality results for Plavnica station – MNE 3

PLAVNICA								
Water layer Parameters	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom
date of sampling	18.04. 2013	18.04. 2013	19.07. 2013	19.07. 2013	22.10. 2013.	22.10. 2013.	7.02. 2014.	7.02. 2014.
Depth, m	-	4.8	-	2,6	-	2,8	-	4.0
Sampling depth, m	0,5	4,0	0,5	1,5	0,5	2.0	0,5	3.0
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
Temperature, °T	16,4	15.0	26,1	25,9	18.0	17,2	8,6	8,5
Transparency, m	3,6	-	2.0	-	2,8	-	2,2	-
Conductivity, µS/cm	255	239	221	217	204	205	255	264
pH	8,23	8,14	8,21	8,35	8,08	8,12	7,64	7,74
Alkalinity, ml HCl	3.3	2,7	2,5	2,4	2,5	-	2,8	2,8
TSS, mg/l	< DL	< DL	0,4	14,2	0,4	0,9	1,9	3.0
DO, mgO2/l	10,77	9,80	9.00	8,70	11,67	9,69	12,10	11,30
O_2 Saturation, %	111	98	112	108	124	101	104	97
COD, mgO2/l	2,14	1,22	2,50	1,70	2,88	1,98	2,10	2,10
BOD_5, mgO2/l	3,08	3,76	1,50	1,30	4,49	2,41	3,70	1,60
NH_3-N, mg/l	0,11	0,07	< DL	0,01	0,02	< DL	0,04	0,02
NO_3-N, mg/l	0,101	0,162	0,016	< DL	0,010	0,050	0,294	0,331
NO_2-N, mg/l	0,003	0,003	< DL	< DL	0,003	0,004	0,005	0,005
TN, mg/l	0.822	0.824	-	-	0,080	0,011	0,570	0,430
o-PO_4-P, µg/l	14	20	17	< DL	14	10	10	43
TP, µg/l	18	25	23	3	15	29	17	56
TOC , mg/l	2.3 0	2.22	-	-	2,33	2,64	1,92	2,16
Cl, mg/l	3,9	3,6	5,8	4,4	3,8	3,5	4,3	3,8
SO_4, mg/l	5,6	6,1	4,9	5.0	5,3	5,5	7,2	7,5
F, mg/l	0.085	0.063	-	-	1,710	1,930	0,070	0,065
Chl-a, mg/l	4,14	-	8,31	-	7,41	-	5,46	-
TC, nfu/100ml	40	150	8	41	54	280	6	4
TFC, nfu/100ml	30	10	0	7	22	260	2	0
HA, nfu/1ml	28	26	37	17	10	91	14	9

Table 3.1.4.: Water quality results for Podhum station – MNE 4

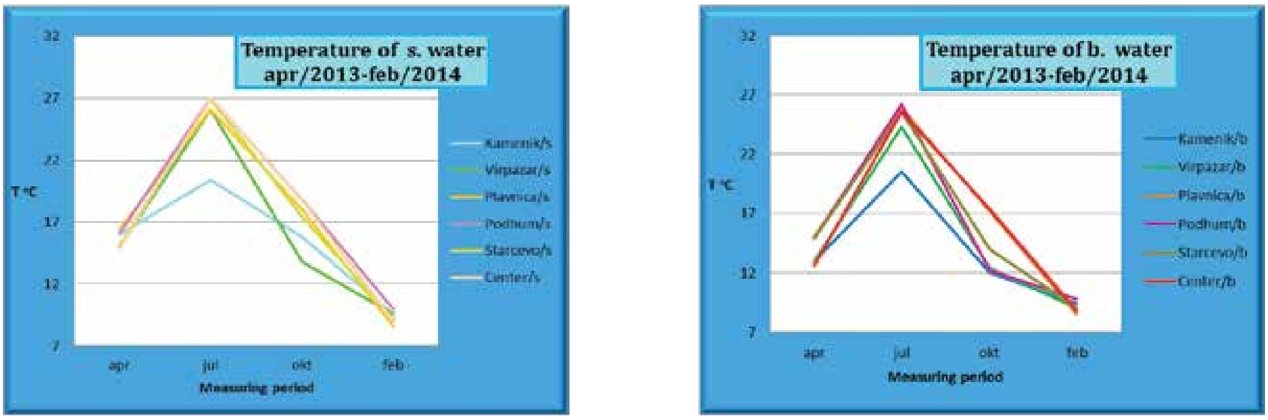
PODHUM								
Water layer Parameters	**Surface**	**Bottom**	**Surface**	**Bottom**	**Surface**	**Bottom**	**Surface**	**Bottom**
date of sampling	17.04. 2013	17.04. 2013	18.07. 2013	18.07. 2013	22.10. 2013.	22.10. 2013.	8.02. 2014.	8.02. 2014.
Depth, m	-	5,5	-	3.0	-	5.0	-	5.0
Sampling depth, m	0,5	4,5	0,5	2,5	0,5	4.0	0,5	4.0
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
Temperature, °T	16,1	14,9	27.0	26,2	18,8	12,2	10.0	9,8
Transparency, m	-	5,5	-	2.7	-	3.4	-	3.1
Conductivity, µS/cm	230	227	210	211	199	227	267	264
pH	8,13	8,03	8,17	8,24	8,37	7,89	7,88	7,88
Alkalinity, ml HCl	2,8	2,4	2,3	2,1	2,5	-	3,1	2,9
TSS, mg/l	< DL	< DL	0,3	0,4	0,3	1,1	0,4	2,5
DO, mgO2/l	10,55	11,19	9,50	9,60	9,90	10,60	12,60	12,50
O_2 Saturation, %	108	111	121	120	107	99	112	110
COD, mgO2/l	0,91	1,33	2,60	2,50	2,58	1,21	1,20	1,40
BOD_5, mgO2/l	1,46	2,23	2,40	2,50	2.00	1,69	1,90	0,90
NH_3-N, mg/l	0,20	< DL	0,03	0,04	< DL	< DL	0,02	0,02
NO_3-N, mg/l	0,347	0,453	0,129	< DL	0,120	0,710	0,589	0,762
NO_2-N, mg/l	0,002	0,002	0,002	0,001	0,002	0,007	0,003	0,002
TN, mg/l	0.742	0.635	-	-	0,170	0,880	1,020	0,730
o-PO_4-P, µg/l	12	2	< DL	< DL	14	14	8	17
TP, µg/l	15	17	23	20	22	16	13	18
TOC , mg/l	1.43	1.12	-	-	1,92	1,05	1,68	1,06
Cl, mg/l	2,7	2,4	4.0	3,4	4,0	3,9	3,1	2,9
SO_4, mg/l	4,8	4,1	4,1	3,5	5,8	0,9	4,7	5.0
F, mg/l	0.060	0.062	-	-	1,940	1,850	0,110	0,035
Chl-a, mg/l	7,70	-	3,90	-	2,34	-	0,84	-
TC, nfu/100ml	12	40	152	77	8	64	0	5
TFC, nfu/100ml	10	2	116	52	3	62	0	2
HA, nfu/1ml	2	7	31	7	6	12	2	6

Table 3.1.5.: Water quality results for Starcevo station – MNE 5

STARCEVO								
Water layer Parameters	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom
date of sampling	17.04. 2013	17.04. 2013	18.07. 2013	18.07. 2013	22.10. 2013.	22.10. 2013.	8.02. 2014.	8.02. 2014.
Depth, m	-	8,5	-	5.8	-	7,5	-	7,5
Sampling depth, m	0,5	7,5	0,5	5,0	0,5	6,5	0,5	6,5
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
Temperature, °T	15,1	14,9	26,9	25,6	17,4	14.0	9,2	9.0
Transparency, m	-	3,7	-	2.5	-	3.3	-	2,4
Conductivity, µS/cm	231	232	207	209	198	242	248	246
pH	8,29	8,19	8,18	8,27	8,29	8,25	8,01	7,93
Alkalinity, ml HCl	2,7	3,2	2,6	2,4	2,3	-	2,8	2,6
TSS, mg/l	< DL	< DL	< DL	0,2	0,2	2,5	1,9	2,3
DO, mgO2/l	11,06	10,68	8,90	8,70	10,33	10,27	12,60	12,60
O_2 Saturation, %	110	106	113	108	109	100	110	109
COD, mgO2/l	0,92	1,16	1,90	2,20	1,75	1,58	1,50	1.00
BOD_5, mgO2/l	2,09	1,86	1.00	1.00	2,47	1,71	2,30	1,60
NH_3-N, mg/l	0,06	0,10	0,02	0,04	0,02	0,03	0,03	0,04
NO_3-N, mg/l	0,225	0,225	< DL	< DL	0,020	0,460	0,373	0,285
NO_2-N, mg/l	0,002	0,001	< DL	0,001	0,003	0,004	0,002	0,002
TN, mg/l	0.721	0.481	-	-	0,170	0,570	0,580	0,620
o-PO_4-P, µg/l	8	9	< DL	< DL	16	19	17	26
TP, µg/l	6	9	23	< DL	17	20	20	27
TOC , mg/l	1.49	1.11	-	-	2,17	1,57	1,48	1,77
Cl, mg/l	2,7	2,9	3,3	3,8	3,4	3,6	2,8	2,6
SO_4, mg/l	5,2	4,7	2,4	4,3	4,5	5.0	5,4	4,7
F, mg/l	0.058	0.072	-	-	0,130	1,480	0,037	0,035
Chl-a, mg/l	7,79	-	3,70	-	3,12	-	4,68	-
TC, nfu/100ml	43	728	0	3	22	70	3	2
TFC, nfu/100ml	16	128	0	1	15	30	2	6
HA, nfu/1ml	19	48	3	2	10	18	2	4

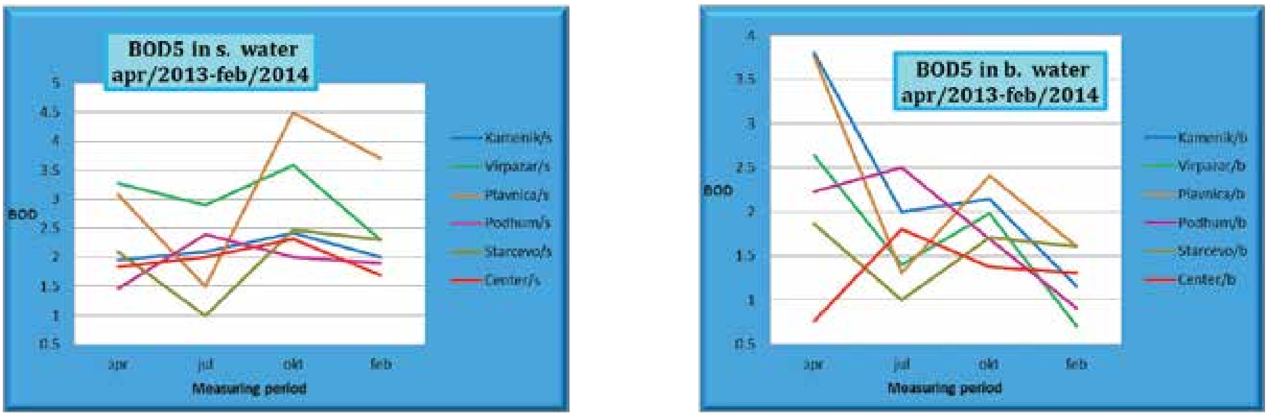
Table 3.1.6.: Water quality results for Center station – MNE 6

CENTER								
Water layer Parameters	Surface	Bottom	Surface	Bottom	Surface	Bottom	Surface	Bottom
date of sampling	17.04. 2013	17.04. 2013	18.07. 2013	18.07. 2013	22.10. 2013.	22.10. 2013.	8.02. 2014.	8.02. 2014.
Depth, m	-	9.0	-	8.0	-	8.0	-	9,5
Sampling depth, m	0,5	8.0	0,5	7.0	0,5	7.0	0,5	8,5
Turbidity, ntu	< DL	< DL	< DL	< DL	< DL	< DL	< DL	< DL
Temperature, °T	14,9	12,6	27.0	25,6	18,0	17,4	9.0	8,8
Transparency, m	-	6,1	-	2.0	-	3.6	-	2,1
Conductivity, µS/cm	223	229	205	207	195	199	238	237
pH	8,20	8,07	8,23	8,28	8,39	8,47	7,98	8,10
Alkalinity, ml HCl	2,8	2,6	2,4	2,3	2,4	-	2,8	2,7
TSS, mg/l	< DL	< DL	< DL	2,6	0,2	2,4	1.0	5,6
DO, mgO2/l	10,63	9,84	8,20	7,90	10,88	10,63	13,10	13.00
O_2 Saturation, %	106	93	104	98	118	112	113	112
COD, mgO2/l	0,95	1,18	1,40	0,80	1,54	1,34	1,50	1,20
BOD_5, mgO2/l	1,84	0,76	2.00	1,80	2,31	1,37	1,70	1,30
NH_3-N, mg/l	0,08	0,10	0,02	0,03	0,02	< DL	0,01	0,01
NO_3-N, mg/l	0,304	0,245	< DL	< DL	< DL	< DL	0,320	0,331
NO_2-N, mg/l	0,002	0,001	0,001	0,001	0,001	0,001	0,003	0,002
TN, mg/l	0.738	0.402	-	-	0,070	0,020	0,450	0,530
o-PO_4-P, µg/l	10	17	7	< DL	11	14	20	13
TP, µg/l	11	28	13	30	14	19	21	20
TOC , mg/l	1.82	1.27	-	-	2,34	1,86	1,58	1,28
Cl, mg/l	2,8	3.0	3,2	3,1	3.0	4,1	3,2	3,5
SO_4, mg/l	5,1	4.0	4,3	3,8	3,6	4.0	5,1	5.0
F, mg/l	0.066	0.069	-	-	0,085	1,650	0,036	0,056
Chl-a, mg/l	2,84	-	8,45	-	3,51	-	1,11	-
TC, nfu/100ml	53	25	7	84	6	160	0	0
TFC, nfu/100ml	40	14	3	76	2	163	0	0
HA, nfu/1ml	6	18	2	9	3	22	2	0



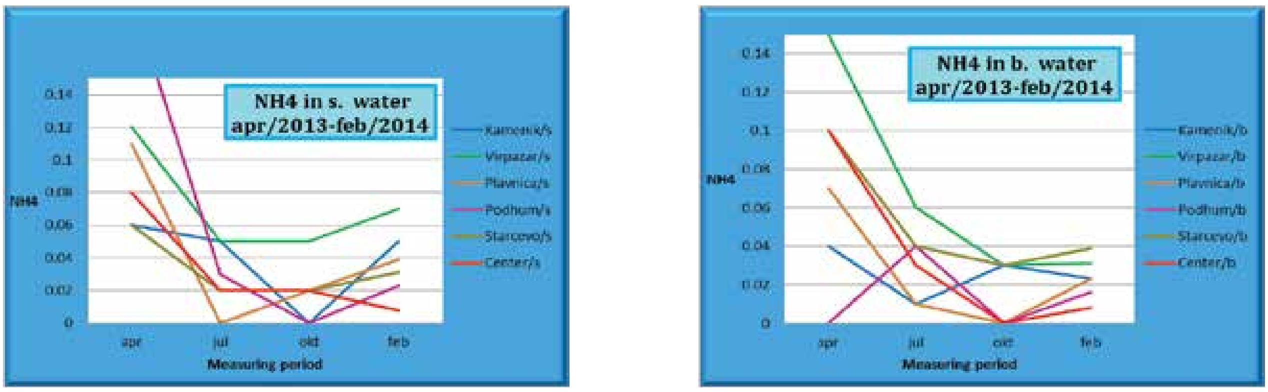
Scheme 1: Temperature April 2013 – February 2014

The differences are being during the summer, when is the maximum. On the end of the period temperatures were equalized and decreased. The highest temperature was at pelagic.



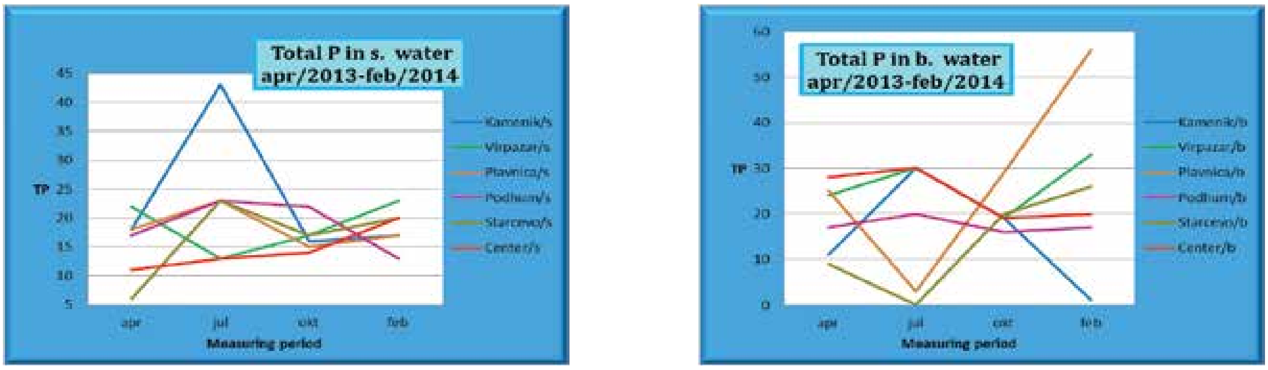
Scheme 2: BOD5 April 2013 – February 2014

There were differences between surface and bottom. Bottom values had max in October at littoral, but not for Center and Podhum. Min values are measured at Starcevo, but max was at Plavnica. Generally the development of the BOD_5 values has been within the normal range.



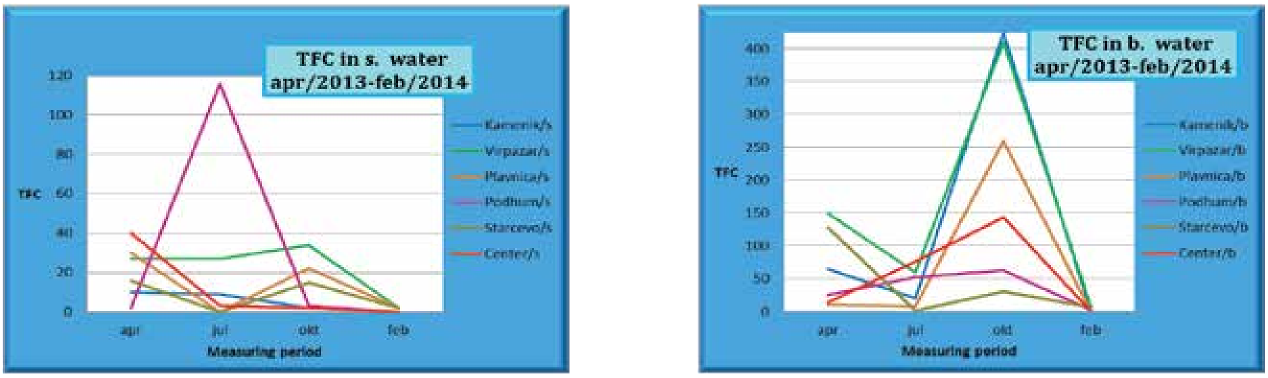
Scheme 3: Ammonia April 2013 – February 2014

Values of ammonia have their minimum in October, except at Virpazar surface and Kamenik bottom layer. Maximal values in April have shown some impact by communal wastewater from the tributaries. Point Virpazar is affected directly by sanitary water from the settlement. Impact from the tributaries has been observed during spring and winter campaigns.



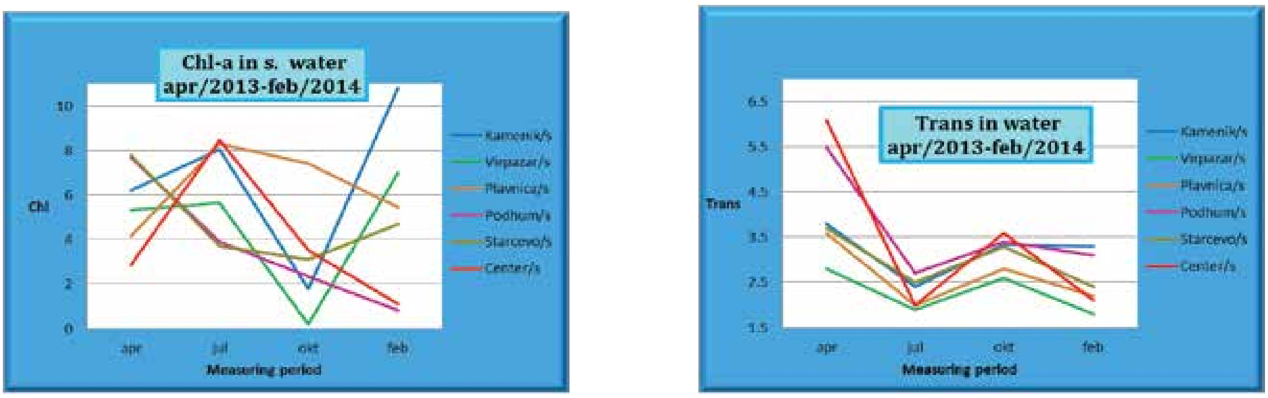
Scheme 4: Total Phosphorus April 2013 – February 2014

Values for Total Phosphorous have shown big alternations between surface and bottom water layer and also during the seasons. Maximum values have been achieved at Kamenik with a peak in summer (surface layer). Except at Kamenik maximum values in summer have been analyzed at Starcevo, Podhum and Virpazar and Plavnica bottom water layer while values at the surface layer has decreased to their minimum). Center and Virpazar has higher values in bottom while Podhum in surface layer. Generally, values of Total Phosphorous are increasing.



Scheme 5: Total Faecal Coliforms April 2013 – February 2014

Total Faecal Coliforms had their maximum in October in the bottom layer. They have decreased to their minimum in July except in Podhum, where was peak in the surface layer. Bottom values were generally higher. However all values haven`t achieved a critical level. The spring values have shown the impact of communal waste water discharges at all sites.



Scheme 6: Chlorophyll-a April 2013 – February 2014

Chlorophyll achieved its maximum in July at Center, Plavnica, Kamenik, Virpazar. Lowest values were analyses in October at Kamenik, Virpazar and Starcevo. Values increased in the winter at Kamenik, Virpazar and Starcevo.

Transparency

Transparency was changing similarly: It is decreased from spring to summer to its minimum, than it increased to autumn, and after, it decreased to winter again. Higher transparency was observed at Podhum, lowest at Virpazar and Plavnica.

Quality of sediment

Table 3.2.: Sediment quality data sampled on 17-18 July 2013

Parameters	M. unit	Kamenik	Virpazar	Plavnica	Podhum	Starcevo	Center
Mercury	μg/kg	116,95	52,25	58,24	58.00	49,50	74,81
Arsenic	mg/kg	0,560	0,486	0,557	3,710	0,623	0,694
Lead	mg/kg	9,26	9,45	6,14	9.00	9,85	6,53
Cadmium	mg/kg	1,20	6,40	3,49	7,31	3,90	3,86
Alumina	g/kg	17,68	12,99	10,56	15,98	13,26	16,78
Organo-chlorine pesticides	μg/kg	< DL	< DL	< DL	< DL	< DL	< DL
Organo-phosphor. pesticides	μg/kg	< DL	< DL	< DL	< DL	< DL	< DL

5.2. Interpretation of the results

Water

The minimal depth of water was at Plavnica (2,6m) and Virpazar (3,0m) in summer. On the other side, the maximal depth was at Starcevo (8,5m – spring) and at Center (9,5m – winter) mainly in spring (April).

The lowest temperature of surface waters was in winter between 8,6°C at Plavnica, and 10,0°C at Podhum. The shallow water at Plavnica is cooled faster. The difference between temperature of surface and bottom waters were within 2°C, without temperature stratification through depth. The maximal surface temperature was in summer, but it not was above 27°C (Podhum and Center).

The transparency (Secchi) was minimal in summer and winter, but maximal in spring (Center 6,1m). Minimal values of transparency ranged in 2,4-3,3m at Kamenik, 1,9-1,8m at Virpazar, 2,0-2,2m at Plavnica, 2,7-3,1m at Podhum, 2,5-2,4m at Starcevo and 2,0-2,1m at Center. These values are caused by bioactive processes (production of biomass) and inflow of polluted waters (sediment) from tributaries.

pH values were minimal at Kamenik and Virpazar, mainly a little bit below 8. Maximal values were observed at Center (surface 8,39, bottom 8,47) in autumn. It is important that at Podhum in autumn, the surface and bottom pH was significantly different: In surface water around 8,37 but in bottom water around 7,89, which could be caused by the discharge of groundwater at the bottom. The hypothesis supported by corresponding water temperature (18,8°C and 12,2°C).

Conductivity was relatively low and corresponds to unpolluted waters, what indicates to absence of ionic substances but presence of organic materials as dominated pollutants. Maximal values were in winter (max 270μS/cm at Virpazar) and the lowest values in autumn, with an absolute minimum of 195μS/cm at Center. The difference of corresponding values between surface and bottom waters was significant at Podhum.

TSS was very low. The increased values at bottom waters, that at times measured, were caused obviously by launched mud from the bottom during sampling.

Dissolved oxygen decreased in spring, passed through minimum in summer, than increased to winter.

Minimal values were at Plavnica, Starcevo and Center. Maximal seasonal difference was at Center: in summer 8,2mg/l; in winter 13,1mg/l.

COD was relatively low. The maximal values were in summer or autumn, in littoral. Minimal values were in pelagic (Center and Starcevo).

BOD_5 was variable within not so large range, from place to place, during seasons and in depth. Maximal values were observed at Plavnica in autumn (4,49mg/l), than at Virpazar. Lowest values were measured at Center, Starcevo and Podhum.

Ammonia was present. The increasing content was mainly in spring in the surface water layer. Maximal values were at Virpazar and Plavnica. Increased value at Podhum measured in spring. The lowest values are typical in autumn at distant sites with deeper water (Center, Starcevo). In conditions with low influence of land-based sources ammonia have been transformed by microbiological activity. Content of ammonia and its spatial distribution corresponded with direct influence of the land sources. When the communal waste waters discharge (the origin of ammonia) was far from lake, the transformation of this substance is done before its coming to lakes` waters.

Nitrates are main nitrogen component. The highest values were in spring or in winter when the land impact was high. The lowest values were in summer, than in autumn, when values sometimes were under detection limit. The amount of nitrates was higher in bottom, in littoral, but at deeper waters` sites in the surface water layer (Center, Starcevo). Maximal values were observed at Podhum, in winter with 0,762mg/l, in bottom water layer.

Nitrites have decrease to their minimum at distant sites as Starcevo and Center but seasonally, in summer. Maximal values were achieved in autumn. Spatial and in depth differences are resultant of land use impact and removal into lake water by microbiological activity. Maximal values were measured in autumn at Podhum (0,007mg/l), Kamenik (0,008mg/l) and Virpazar (0,009mg/l), all in bottom water.

The key factor for eutrophication, phosphorus was significantly present. Maximal values of total phosphorus were at littoral stations Kamenik, Virpazar and Plavnica, but minimal values were at Starcevo and Center. Spatial, in depth and seasonal changing of this parameter were very variable,

Table 4.: National MAV of the surface waters

Parameter	Measurement units	MAV for A2 class
Temperature	^{0}C	30
Turbidity	NTU	5
Conductivity	µS/cm	600
Total Suspended Solids	mg/l	20
pH	-	6.5-8.5
DO	mg/l	-
Saturation O_2	%	80-120
BOD_5	mg/l	4
COD ($KMnO_4$)	mg/l	8
Ammonium ion	mgN/l	0.04
Nitrates	mgN/l	5.65
Nitrites	mgN/l	0.002
Orto-Phosphates	mgP/l	0.03
Total phosphorus	-	-
Fluorides	mg/l	1.5
Sulfates	mg/l	50
Chlorides	mg/l	40
Chlorophyll a	mg/l	-
TOC	mg/l	2
Total coliforms 37°C	Nfu in 100ml	5000
Faecal coliforms	Nfu in 100ml	2000

according opposite processes of its inflow and removal. The concentration of TP is ranged: at Kamenik 1-43µgP/l (July); at at Virpazar 13-33µgP/l (February); at Plavnica 3-56µgP/l (February); at Podhum 13-23µgP/l (July); at Starcevo <DL-26µgP/l (February); at Center 2-30µgP/l (July).

Chlorophyll-a was very variable. The values ranged from 0,2mg/l at Virpazar to 10,08mg/l at Kamenik. The minimal values were mainly in autumn, but at Podhum and Center in winter and at Plavnica in spring. Maximal values occurred in different seasons at different places, which indicate very complex phytoplankton activity.

Abundance of faecal originated bacteria was moderate, with peaks mainly in autumn (but at Podhum in summer and at Starcevo in spring), and in bottom waters (except at Podhum). Spatial distribution is shown the maximal values at Kamenik. The values decreased in following order: Virpazar, Plavnica, Podhum, Starcevo and Center. The extreme peak at Starcevo in spring should take with reserve. Number of Heterotrophic aerobic bacteria was generally low and corresponded to faecal bacteria values. It should note the illogical results (``0``) of its presence at Center in winter.

The requested class of water quality of Skadar Lake has classified as A2, according Regulation of surface and ground waters` classification and categorisation ("Off. Gaz. of Montenegro", No.2/07). The maximum allowed values (MAV) related to this class, presented in table below.

The status of compliance of water quality with related national legislative was as follow.

The most of measured parameters (classified by national regulation) were in requested A2 class. Only several parameters were above this class. Ammonia was always out of A2 at Virpazar in surface, but mainly in bottom water. The values at Kamenik mainly were out of A2 class, but at all other sites, ammonia content was out of A2 in spring only.

Nitrites were most often out of A2 at Plavnica (always, except in summer), than at Kamenik and Virpazar. At other sites, the content of nitrites exceeded A2 in autumn only.

Fluorides exceeded the A2 only in autumn, at all stations, sometimes through water column, or in the bottom.

TOC was mainly above A2 at Plavnica. This parameter at Kamenik, Starcevo and Center, was out of A2 only in autumn, but at Virpazar, in spring.

Orto-phosphorus was above A2 only at Plavnica (winter) and at Center (summer).

Sediment

Because missing of the related national regulations for sediment, the results are compared with Dutch standards - allowed limits (see Table 4.).

Table 5.: MAV and HRV for hazardous and harmful substances in the soil (Dutch Standards, 2000, 2009)

Metal	m. unit	MAV*	HRV*
Mercury	mg/kg	0,3	10
Arsenic	mg/kg	29.55	55
Lead	mg/kg	85	530
Cadmium	mg/kg	0.8	12

** MAV – maximal approved values*
HRV – high risk values

The results only for cadmium were above MAV at all stations (1,5-9,1 times). Content of mercury was 0,2-0,4 times below, arsenic 0,02-0,1 times, lead 0,07-0,1 times. Alumina had very high values, with order of magnitude in grams. This metal contributed in geological structure, but founded increased values, could be contribution from land based sources, first of all from Aluminium Plant in Podgorica. The content of selected metals has been some higher than in 90-ies of last century [Royal Haskoning 2006], but it has been lower than in 2005 [Sundic and all 2012].

Mercury was minimal at Virpazar. The other results were similar, but the maximum at Kamenik was about 2 times higher. The results were similar, but it is stand out the maximum at Podhum. Lead is increased at Kamenik, Virpazar, Podhum and Starcevo. Cadmium was extremely minimal at Kamenik, but it is increased at Podhum (max) and Virpazar. Alumina was very similar at all sites. The maximum was at Kamenik, but minimum at Plavnica.

Generally, the biggest amount of heavy metals in sediment was found at Podhum and Kamenik. In the first site it is probably the consequence of groundwaters impact, but in second place, influence of the river Crnojevica and the right branch of the river Moraca (maybe of leaching waters from Cetinje dump, too).

The lowest content of heavy metals in sediment is measured at Plavnica, which is out of direct sources influence, also out of main stream (and mixture) of waters in the Lake.

6. Conclusions and Recommendations

- Water and sediment quality have been investigated within transboundary coordinated campaigns for the first time by Albanian and Montenegrin partner institutions.
- The lessons learned and experience, as well as provided equipment and applied measuring program which were result in capacity building of our responsible institution, ensure the possibility to do the future monitoring of physical-chemical and microbiological parameters of Skadar Lake water body according to the WFD requirements.
- The results of the investigation are the base for the evaluation of biological investigations which have been carried out at the same time to assess the ecological status of the water bodies of Skadar Lake.
- On the base of the results the monitoring program according article 8 WFD could be established including the biological elements.

Due to fulfill the overall goals, it is recommended to follow up the project activities by extending the project terms with new, project purposed outlines.

- Characterization of hydro-morphological conditions
- Verification of the initial characterization including the delineation of the water bodies at the Lake on the base of future monitoring program
- Extend the monitoring program to tributaries at their mouth areas, as well as groundwater of Zeta Plane, at characteristic points. This monitoring has to be in the same time as water quality monitoring of the Lake.
- Determination of the pollution load discharged to the lake from different land sources.

Literature

Royal Haskoning, 2006. (Keukelaar, F., De Goffau, A., Pradhan, T., Sutmuller, G., Mišurović, A., Ivanović, S., Uskoković, B., Hetoja, A., Haxhimihali, E., Prifti, A., Kapidani, E., Kashta, L. & Gulan, A.). Lake Shkoder transboundary diagnostics analysis Albania & Montenegro. Project number 9P6515, Podgorica, 111 p.

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Decree on categorization and classification of surface and ground waters in Montenegro, ``Off. Gaz. of Montenegro``, No.2/07.

Phytoplankton of Skadar Lake
-Final Report-

Dr Jelena Rakocevic

University of Montenegro
Faculty of Natural Sciences, Biology Department

Data from Albania from
Dr Marash Rakaj

University of Shkodra

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Contents

1. Introduction

The central tenet of the Water Framework Directive (2000/60/EC) is that key communities, or Biological Quality Elements (BQEs), are used to assess the water quality of lakes, rivers and coastal/transitional waters. Phytoplankton has been identified as a key BQE to assess the ecological quality of lakes and is widely used as a water quality indicator because of its high species differentiation and sensitivity to environmental changes (i.e. affected by physical, chemical and biological factors). Phytoplankton communities respond sensitively to eutrophication pressures and are considered the most direct of all biological quality elements. Phytoplankton as primary producers have a primary role in the food web and an important influence on other organisms. Phytoplankton is increasingly used to monitor the ecological quality and health of the water environment and to measure the effectiveness of management or restoration programs or regulatory actions.

As parameters to be studied, the WFD (Annex V) prescribes phytoplankton abundance, composition, biomass and the frequency and intensity of blooms. All these parameters are considered to undergo degradation along pressure gradients and the extent of this degradation can be translated into WFD normative definitions.

2. Methods

The phytoplankton investigation of Skadar Lake comprised the assessment of the following parameters:

- Phytoplankton composition - list of all present phytoplankton taxa
- Phytoplankton abundance – number of individuals per liter of water (cells/l)
- Diversity of phytoplankton community - by calculating diversity index (Shannon-Wiener index)
- Bloom metric - Pielou's evenness index (shows how equal the community is numerically)
- Bloom metric - Cyanobacterial abundance
- Phytoplankton biomass – chlorophyll *a* concentration
- Trophic State Index (TSI)
- Autecology of dominant species (gives information about general water quality, type of the lake (deep shallow, glacial – lowland etc.), trophic level (eutrophication), toxicity (blue-greens), and pH/Ca –Desmids etc.)

Sampling points. Phytoplankton investigation comprised sampling from six sampling points at the Montenegrin and three sampling points at the Albanian part of Skadar Lake. Sampling points in Montenegro were Kamenik (northwestern part of Skadar Lake – Vucko blato), **Plavnica** and **Podhum** (on the north coast of the lake), **Starcevo** and **Virpazar** (on the south coast of the lake) and **Center** (pelagial). The Albanian sampling points were **Sterbeq**, **Zogaj** and **Shiroke**, which are all located in the littoral zone of the lake (Fig.1).

Sampling campaigns. In most lakes, phytoplankton reaches several well distinguishable phases in any individual year, among which spring diatom phase and late summer phase are usually the most distinguishable in majority of lakes.

Spring diatom development and composition reflect characteristic features of different lakes including their trophic state. The timing is strongly dependent on the presence/absence and extension of the winter ice cover. The main problem about determining the optimum sampling time (date) are the unexpectedness and relative shortness of this phase. For Skadar Lake, it is usually the end of March/beginning of April (it varies from year to year, especially considering complete absence of winter ice cover). After the collapse of the spring diatom bloom, the clear water phase appears even though it is often not so evident in polymictic lakes such as Skadar Lake.

The late summer community integrates the preceding successional events (including the onset of stratification and disturbances) and is strongly determined by the competitive arena that any given lake type offers. Besides, during this period of the year cyanobacterial blooms are most likely to occur. Samples at Skadar Lake should therefore be taken preferably between August 15 and September 1.

For these reasons, phytoplankton samples were taken during two sampling campaigns in spring (April 25) and summer (August 28), respectively, in both the Montenegrin and the Albanian part of the lake.

Sampling. For the qualitative analyses of phytoplankton, samples were taken using plankton net No. 25 and fixed with Lugol's Iodine. Samples for quantitative analyses of phytoplankton were taken using a hydrobiological Ruttner sampling bottle. Previous phytoplankton investigations showed a uniform distribution of phytoplankton from the surface to the bottom owing to the shallowness of Skadar Lake and frequent mixing events. For that reason, all samples were taken 30 cm under the water surface. Each sample was clearly labeled indicating sampling site, date, time and sampling depth. Sampling bottles were transported to the laboratory in a cooler box.

Analyses. Identification of phytoplankton taxa was done according to Bourrely (1970, 1981), Komarek & Anagnostidis (1999), Krammer & Lange-Bertalot (1986. 1988, 1990), Pestalozzi, Kramer & Fott (1983), Popovsky & Pfiester (1990). Quantitative analyses (enumeration of individuals per liter) were done according to Utermöhl (1958) using an inverted microscope Axiovert 25. A low magnification (40x or 100x) whole chamber count, two intermediate magnification (200x or 250x) transect counts and 50-100 field of view counts at high magnification (400x) were performed. Chlorophyll *a* analyses were made according to ISO 10260 (1992) and the concentration of this pigment was used as an indirect measure of phytoplankton biomass. The trophic state index (TSI) was calculated according to Carlson (1977), the evenness index according to Pielou (1969) and the diversity index according to Shannon-Wiener (1949).

Figure 1. Sampling points for phytoplankton investigation in Skadar Lake

3. Existing data and gaps

Phytoplankton at the Montenegrin part of Skadar Lake was first studied at the end of fifties of the last century and then periodically till the end of the eighties. The majority of these investigations were qualitative in nature (taxonomic inventories). Quantitative analyses of phytoplankton were done occasionally, focusing exclusively on phytoplankton abundance and rarely covering more than one season. One of the most consistent effects of eutrophication on phytoplankton (both natural and anthropogenic) is a shift in species composition. However, data on changes in biodiversity and trophic conditions are largely missing because of the lack of repeated investigations. After a pause of almost three decades, investigations of phytoplankton in Skadar Lake continued in the first decade of the century (Rakocevic & Hollert, 2005). These studies investigated spatial and temporal variation in phytoplankton composition and abundance at Skadar Lake in order to improve the knowledge on phytoplankton succession patterns in shallow subtropical lakes.

4. Results

4.1 Phytoplankton composition

Montenegro

During the spring sampling campaign, 65 species of phytoplankton from 4 divisions were identified: Chlorophyta – 29 species, Bacillariophyta – 28 species, Pyrrophyta – 6 species and Chrysophyta – 2 species (Fig.2).

During the summer sampling campaign, 56 species of phytoplankton from 6 divisions were identified: Chlorophyta – 22 species, Bacillariophyta – 17 species, Cyanophyta – 7 species, Pyrrophyta – 5 species, Chrysophyta – 2 species and Euglenophyta – 2 species (Fig.3). Summer phytoplankton communities contained 2 additional algal divisions (Euglenophyta and Cyanophyta) but a lower proportion of Bacillariophyta, in comparison with spring communities.

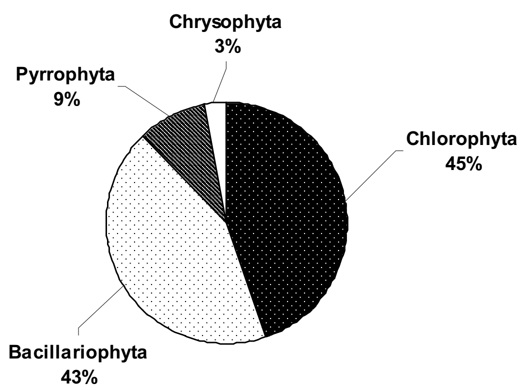

Figure 2. Percentage of algal divisions of the total number of identified species at the Montenegrin part of Skadar Lake in spring (April 2013)

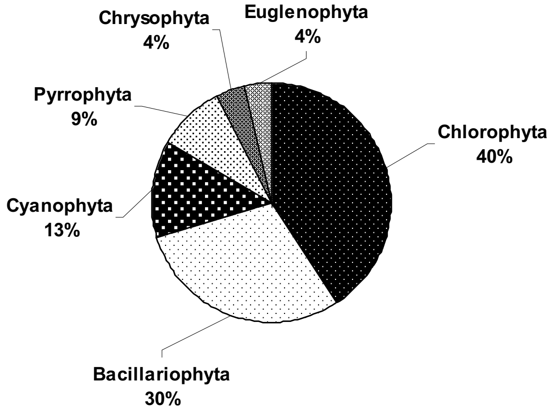

Figure 3. Percentage of algal divisions of the total number of identified species at the Montenegrin part of Skadar Lake in summer (August 2013)

Species number was lowest at the center of the lake in both sampling campaigns (Tab. 1). This was expected, considering that the sampling point was located in the deepest part of the lake, representing the only typically pelagic point and characterized by phytoplankton communities consisting mainly of euplanktonic species. Other sampling points are shallower and their phytoplankton communities contain littoral species resuspended from the bottom, shore or macrophytes. The summer sampling campaign yielded higher species numbers than the spring campaign, with a higher proportion of Chlorophyta.

Albania

During the spring sampling campaign, 47 species from 5 phytoplankton divisions were identified: Bacillariophyta – 30 species, Chlorophyta – 6 species, Chrysophyta – five species, Pyrrophyta – 4 species and Cyanophyta – 2 species (Fig.4).

During the summer sampling campaign, 91 species belonging to 6 divisions were identified: Bacillariophyta – 45 species, Chlorophyta – 28 species, Cyanophyta – 12 species, Pyrrophyta – 4 species and Chrysophyta – 2 species (Fig.5).

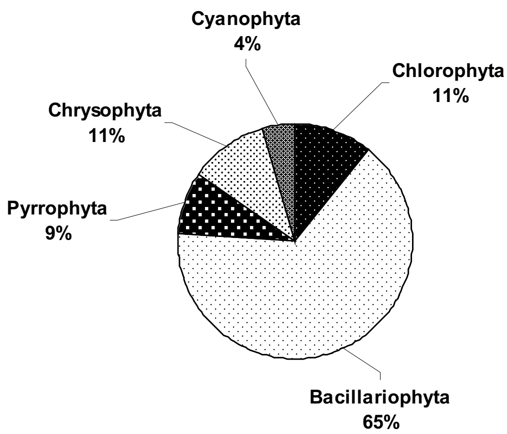

Figure 4. Percentage of algal divisions of the total number of identified species at the Albanian part of Skadar Lake in spring (April 2013)

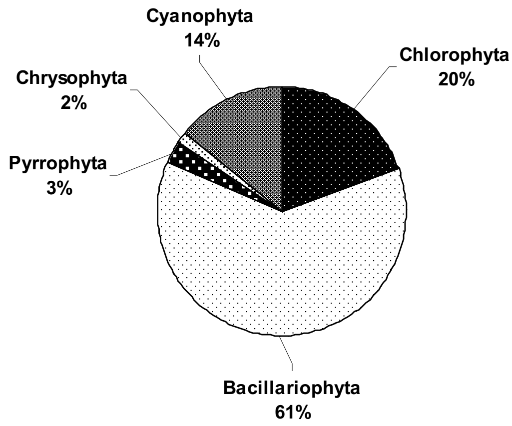

Figure 5. Percentage of algal divisions of the total number of identified species at the Albanian part of Skadar Lake in summer (August 2013)

In both sampling campaigns, the highest phytoplankton species number was recorded at Zogaj and the lowest at Sterbeq. Species diversity was higher in summer than in spring and showed a higher proportion of Chlorophyta.

4.2 Phytoplankton indices

Montenegro

During the spring sampling campaign, **diversity** and **evenness** (Tab. 1) were highest at the center of the lake (pelagial) and lowest at Podhum where *Dynobryon bavaricum* was by far the most dominant species, accounting for 90% of the total phytoplankton abundance (spring bloom). Diversity and evenness were therefore lowest at this locality.

The opposite was true in summer when both indices were lowest in the pelagial zone and highest at Podhum (Tab. 1). The diatom species *Aulacosira italica* accounted for 56% of the total phytoplankton abundance, leading to low diversity and evenness in summer.

Table 1. Species number, diversity index and evenness index at 6 sampling points at the Montenegrin part of Skadar Lake in spring (I) and summer (II)

	Kamenik		Virpazar		Plavnica		Podhum		Pelagial		Starcevo	
	I	II	I	II	I	II	I	II	I	II	I	II
Species number	37	41	29	39	26	35	22	40	19	20	42	31
Diversity index	2.33	3.99	2.10	3.94	2.23	3.64	1.48	4.49	2.91	2.13	2,80	3.01
Evenness index	0.45	0.75	0.42	0.74	0.47	0.74	0.34	0.86	0.68	0.50	0.52	0.63

Albania

During the spring sampling campaign (Table 2), the highest species number was found at Zogaj (37) and Shiroke (35) and the lowest at Sterbeq (26). **Diversity** and **evenness** were highest at Zogaj and lowest at Sterbeq. The high values at Zogaj were due to: 1. low abundance of phytoplankton at this locality and 2. more or less uniform distribution of species abundances. The phytoplankton at Zogaj and Shiroke was dominated by Chrysophyceae, mainly *Dinobryon bavaricum* and to a lesser extent *Dinobryon divergens*. The second most dominant group were diatoms, represented mainly by *Cyclotella glomerata*.

During the summer sampling campaign (Table 2), the highest species number was recorded at Zogaj (78) and Shiroke (71) and the lowest at Sterbeq (66). In summer, species diversity was generally higher in Albania than in Montenegro. Similar to the spring sampling campaign, **diversity** and **evenness** were highest at Zogaj and lowest at Sterbeq and Shiroke. High diversity and evenness values at Zogaj were due to a uniform distribution of species abundances. The phytoplankton communities were dominated by diatoms at all three stations, the majority belonging to the two genera *Aulacosira* and *Cyclotella*.

Table 2. Species number, diversity index and evenness index at 3 sampling points at the Albanian part of Skadar Lake in spring (I) and summer (II)

	Sterbeq		Zogaj		Shiroke	
	I	II	I	II	I	II
Species number	26	66	37	78	35	71
Diversity index	2.06	2.63	2.61	3.39	2.38	2.70
Eveness index	0.54	0.58	0.68	0.75	0.62	0.60

4.3. Phytoplankton abundance

Montenegro

During the spring sampling campaign, the highest **total abundance** of phytoplankton (Tab. 3.) was found at Kamenik and Virpazar and the lowest at the center of the lake (pelagial). Kamenik and the sampling points located at the southern part of the lake (Virpazar and to a lesser extent Starcevo) were dominated by Bacillariophyta, mostly by *Fragilaria crotonensis*. At the opposite lake side further up north (Plavnica and Podhum), phytoplankton communities were dominated by Chrysophyta (*Dynobryon bavaricum* and *D. divergens*). At the pelagial zone, Bacillariophyta and Chrysophyta were about equally abundant, with *Dinobryon bavaricum* (Chrysophyta) being the dominant species. *Asterionella formosa* and small *Cyclotella* species were the most abundant diatoms at this locality.

During the summer sampling campaign, the highest **total abundance** of phytoplankton was recorded at Kamenik and Starcevo and the lowest at Podhum (Tab. 4).

Table 3. Total phytoplankton abundance (cells/l) and abundance of different divisions of algae at the Montenegrin part of Skadar Lake in spring (April 2013)

	Kamenik	Virpazar	Plavnica	Podhum	Pelagial	Starcevo
Total abundance	$7.0x10^6$	$6.6x10^6$	$4.4x10^6$	$2.0x10^6$	$6.5x10^5$	$3.5x10^6$
Bacillariophyta	$6.1x10^6$	$5.6x10^6$	$7.0x10^5$	$2.1x10^5$	$3.1x10^5$	$1.9x10^6$
Chrysophyta	$6.2x10^5$	$9.4x10^5$	$3.6x10^6$	$1.8x10^6$	$3.1x10^5$	$1.4x10^6$
Chlorophyta	$1.2x10^5$	$1.0x10^5$	$1.8x10^5$	$1.3x10^4$	$3x10^4$	$2.3x10^5$
Pyrrophyta	$6.3x10^3$	$5.1x10^3$	$3.0x10^3$	$4.0x10^3$	$1.0x10^3$	$4.0x10^3$

Sampling points at the northern part – Plavnica and Podhum – were dominated by small centric euplanktonic Bacillariophyta, mainly *Cyclotella* species. Bacillariophyta also dominated in the more open parts of the lake at the center and at Starcevo, the most abundant species being *Aulacoseira italica*. Chlorophyta were represented at all these localities by similar species – mostly Chlorococcales, and Cyanophyta by *Merismopaedia* species. At Virpazar and Kamenik, the filamentous green alga

Planktonema lauterbornii and the pennate diatom *Fragilaria ulna* var. *acus* dominated, while Cyanophyta were represented mostly by the filamentous alga *Anabaena variabilis*.

Table 4. Total phytoplankton abundance (cells/l) and abundance of different divisions of algae at the Montenegrin part of Skadar Lake in summer (August 2013)

	Kamenik	Virpazar	Plavnica	Podhum	Pelagial	Starcevo
Total abundance	$4.0x10^6$	$3.2x10^6$	$1.85x10^6$	$5.5x10^5$	$3.5x10^6$	$3.9x10^6$
Bacillariophyta	$1.5x10^6$	$1.5x10^6$	$9.3x10^5$	$3.7x10^5$	$3.0x10^6$	$3.5x10^6$
Chrysophyta	$9.2x10^4$	$3.5x10^4$	$1.2x10^4$	$1.1x10^4$	$1.0x10^3$	$1.0x10^3$
Chlorophyta	$1.6x10^6$	$9.1x10^5$	$2.8x10^5$	$2.5x10^5$	$1.2x10^5$	$4x10^5$
Pyrrophyta	$1.8x10^5$	$4.2x10^5$	$2.1x10^5$	$1.4x10^4$	$1.0x10^3$	$1.0x10^3$
Euglenophyta	$1.2x10^5$	$1.0x10^4$	$2.3x10^5$	$6.0x10^3$	$1.0x10^3$	$1.0x10^3$
Cyanophyta	$5.3x10^5$	$1.7x10^5$	$6.4x10^5$	$2.1x10^4$	$5.3x10^4$	$7x10^4$

Albania

The highest **total abundance** of phytoplankton (Tab. 5) in spring was recorded at Zogaj and Shiroke and the lowest at Sterbeq. *Dinobryon bavaricum* and *Dinobryon divergens* (Chrysophyta) were the dominant species at all three sampling points, followed by the second most abundant algal division Bacillariophyta, represented mainly by *Cyclotella glomerata*, while other phytoplankton groups were present at very low abundance.

Table 5. Total phytoplankton abundance (cells/l) and abundance of different divisions of algae at the Albanian part of Skadar Lake in spring (April 2013)

	Sterbeq	Zogaj	Shiroke
Total abundance	$4.1x10^4$	$5.6x10^4$	$5.3x10^4$
Chrysophyta	$2.8x10^4$	$1.7x10^4$	$2.1x10^4$
Bacillariophyta	$1.2x10^3$	$3.7x10^3$	$2.9x10^3$
Pyrrophyta	$6.1x10^2$	$9.8x10^2$	$1.2x10^3$
Chlorophyta	$3.4x10^2$	$6.2x10^2$	$6.9x10^2$
Cyanophyta	-	-	$2.0x10^2$

During the summer sampling campaign, the highest **total abundance** of phytoplankton (Table 6) was recorded at Shiroke and the lowest at Zogaj, which correlated well with water transparency (highest value observed at Zogaj, see Table 11) and dissolved oxygen. Diatoms (Bacillariophyta) dominated the phytoplankton communities at all three stations represented mainly by *Cyclotella ocellata, C. distinguenda* and *Alcoseira italica*. Chlorophyta were represented with similar species – mostly Chlorococcales, while *Merismopaedia* species dominated among Cyanophyta, especially at Shiroke station. *Ceratium hirudinella* (Pyrrophyta) was most numerous at Zogaj and Shiroke. *Fragilaria ulna* was well represented at Shiroke and Sterbeq (in shallow water). The large forms of *Pediastrum, Ceratium, Merismopedia, Microcystis* and filamentous forms of *Oscillatoria, Planktolyngbya* and *Pseudoanabaena* were observed at high density mostly at Shiroke and Zogaj (mesotrophic condition) and to a lesser extent at Sterbeq (oligo-mesotrophic condition). The substrata at Zogaj and Sterbeq are characterized by rock and/or gravel and are only sparsely vegetated compared to Shiroke with muddy substrata and well developed vegetation.

Table 6. Total phytoplankton abundance (cells/l) and abundance of different divisions of algae at the Albanian part of Skadar Lake in summer (August 2013)

	Sterbeq	Zogaj	Shiroke
Total abundance	$3.1x10^5$	$3.2x10^5$	$3.4x10^5$
Chrysophyta	$9.1x10^3$	$8.3x10^3$	$7.8x10^3$
Bacillariophyta	$1.5x10^5$	$1.5x10^5$	$1.7x10^5$
Pyrrophyta	$2.3x10^4$	$2.0x10^4$	$2.6x10^4$
Chlorophyta	$5.3x10^4$	$4.6x10^4$	$6.0x10^4$
Cyanophyta	$5.7x10^4$	$6.1x10^4$	$7.3x10^4$

Autecology of dominant species. The following table provides an overview of autecological characteristics of the most abundant phytoplankton species recorded at Skadar Lake (April and August 2013, Montenegro and Albania):

<table>
<tr><th></th><th>Algal division:</th><th>Species:</th><th>Autecology:</th></tr>
<tr><td rowspan="2">APRIL</td><td>Bacillariophyta</td><td>Fragilaria crotonensis
Asterionella formosa</td><td>Total phosphorus (TP) optima in range of meso- to eutrophic waters. These species are typical for spring blooms particularly in nutrient-enriched lakes.</td></tr>
<tr><td>Chrysophyta</td><td>Dynobryon bavaricum
Dynobryon divergens</td><td>Mixotrophic species feeding on bacteria in case of nutrient deficiency (especially phosphorus deficiency); mainly found in oligotrophic, phosphorus-poor waters.</td></tr>
<tr><td rowspan="4">AUGUST</td><td>Bacillariophyta</td><td>Fragilaria ulna var. acus
Aulacoseira italica
Cyclotella species</td><td>Fast-growing diatoms mostly found in shallow, nutrient-enriched, well-ventilated waters prone to become turbid.</td></tr>
<tr><td>Chlorophyta</td><td>Planktonema lauterbornii
Chlorococcales</td><td>Usually dominate phytoplankton assemblages in eutrophic lakes.</td></tr>
<tr><td rowspan="2">Cyanophyta</td><td>Anabaena variabilis
(Montenegro)</td><td>Typically found in late summer-autumn plankton under warm water conditions; capable of storing phosphorus and fixing atmospheric nitrogen</td></tr>
<tr><td>Merismopaedia
(Albania)</td><td>Late summer-autumn plankton under warm water conditions in stratified meso- to eutrophic lakes</td></tr>
</table>

4.4. Chlorophyll *a* concentration and Trophic State Index (TSI)

Montenegro

Trophic State Index (TSI) was calculated based on values of transparency (Secchi Disc – SD) and chlorophyll *a* concentrations (Chl) and the results for the spring sampling campaign (April, 2013) are presented in Table 8.

Table 8. Water transparency, chlorophyll *a* concentration and values of two TSI-indices at six sampling stations in Montenegrin part of Skadar Lake (April, 2013)

Stations	Transparency (m)	Chlorophyll *a* (µg/l)	Trophic State Index (Chl)	Trophic State Index (SD)
Kamenik	3.8	6,2	48	67
Virpazar	2.8	5,3	47	72
Plavnica	3.6	4,1	44	68
Podhum	5.5	7,7	50	62
Pelagial	6.1	2.8	40	60
Starcevo	3.7	8.0	51	68

The lowest concentration of chlorophyll *a* and the lowest TSI's (based on chlorophyll *a* and transparency) in spring were found at the pelagial zone which correlates well with the low phytoplankton abundance at this point. The TSI based on chlorophyll *a* indicates mesotrophic conditions. The higher TSI based on transparency (i.e. eutrophic conditions) indicates that water turbidity was not entirely caused by phytoplankton but also by other particles suspended in the lake water (water circulation connected with frequent winds). The average value of TSI (Chl) in spring indicates mesotrophic conditions of the lake water.

Owing to lower phytoplankton abundance the average TSI (Chl) value was lower in summer than in spring. Values for different localities are presented in Table 9. The highest values of chlorophyll *a* concentration and TSI were observed at Kamenik and Virpazar (eutrophic conditions) and the lowest values at the pelagial zone and Podhum (oligo-mesotrophic conditions). The average value of TSI (Chl) in August indicates mesotrophic conditions of the lake water.

Table 9. Water transparency, chlorophyll *a* concentration and two TSI-indices at six sampling stations at the Montenegrin part of Skadar Lake in summer (August 2013)

Stations	Transparency (m)	Chlorophyll a (µg/l)	Trophic State Index (Chl)	Trophic State Index (SD)
Kamenik	2.7	12.5	56	71
Virpazar	2.3	8.7	51	74
Plavnica	2.0	6.9	49	78
Podhum	3.0	2.8	37	70
Pelagial	2.4	2.1	36	72
Starcevo	2.0	3.3	43	78

Albania

During the spring sampling campaign, the observed values of chlorophyll *a* and TSI (Chl) clearly indicate oligotrophic conditions at all three sampling points (Tab. 10). The highest values of TSI (Chl) and the lowest values of TSI (SD) were observed at Zogaj station. This paradox was presumably due to unstable water conditions at the littoral part of the lake. The high TSI (Chl) in Zogaj was well correlated with the chlorophyll *a* concentration, while the lower value of TSI (SD) compared to Shiroke and especially Sterbeq was due to unstable water conditions at the latter two sampling points. TSI (Chl) and phytoplankton abundance were correlated at all three stations. The poor species richness and low biomass (chlorophyll *a*) was probably the result of high water level and unstable water conditions at the time of sampling.

Table 10. Water transparency, chlorophyll *a* concentration and TSI at the Albanian part of Skadar Lake in spring (April 2013)

Stations	Transparency (m)	Chlorophyll a (μg/l)	Trophic State Index (Chl)	Trophic State Index (SD)
Sterbeq	2.75	1.1	31	45
Zogaj	3.62	1.7	36	41
Shiroke	2.87	1.3	33	47

During the summer sampling campaign, the values of chlorophyll *a*, TSI (SD) and TSI (Chl) indicate mesotrophic conditions at all three sampling points (Tab.11). Small differences in TSI values between the three sampling points showed similar water conditions. TS indices (transparency and chlorophyll *a* concentration) corresponded well correlation with phytoplankton abundance at all sampling stations.

Table 11. Water transparency, chlorophyll *a* concentration and values of two TS indices at the Albanian part of Skadar Lake in summer (August 2013)

Stations	Transparency (m)	Chlorophyll a (μg/l)	Trophic State Index (Chl)	Trophic State Index (SD)
Sterbeq	2.3	3.1	42	48
Zogaj	2.6	2.9	41	46
Shiroke	2.1	3.4	43	49

5. Conclusions

Considering the taxonomic composition and abundance of phytoplankton assemblages at Skadar Lake as well as autecological characteristics of dominant species, it can be concluded:

- Kamenik and Virpazar (Montenegro) showed the highest trophic state of all sampling sites, showing **mesotrophic** conditions in spring and typical **eutrophic** conditions in summer. Kamenik is located at the Vucko mud – a part of the lake with very limited hydrologic communication with the main water mass and under influence of Crnojevica River (fish-plant, communal water from the city of Cetinje) and the right arm of Moraca River. The sampling point Virpazar is under human influence of the Virpazar settlement and Virpazarska River.
- The pelagic sampling point (Montenegro) and the Starcevo and Plavnica sampling points showed **oligo-mesotrophic** conditions in spring and **slightly eutrophic** conditions in summer.
- The sampling site at the northern shore – Podhum (Montenegro) showed the lowest trophic state of all sites investigated in Montenegro. Conditions were **oligotrophic** in spring and **mesotrophic** in summer. At this somewhat isolated sampling point nutrient loads are generally small, originating mostly from fishing activities. The observed trophic level was therefore expected.
- The Albanian sampling stations (Sterbeq, Zogaj and Shiroke) have similar trophic conditions

– **oligotrophic** in spring and **mesotrophic** in summer. The observed tendency towards eutrophication in summer is mainly due to human activities such as urbanization accompanied by the discharge of sewage water directly into the lake (Zogaj, Shiroke), fishing (especially at Zogaj and Shiroke), agriculture with the use of chemical fertilizers (especially at Sterbeq), animal husbandry (Zogaj, Sterbeq) and deforestation.

- The results of phytoplankton analyses at the Albanian side of Skadar Lake were most similar to those observed at Podhum in Montenegro, which is located close to the border between two countries.
- The results from both the Montenegrin and Albanian part sides of Skadar Lake provide evidence of the following:

1) Increase in trophic state from early spring to end of summer

2) Decrease of trophic state from north-west to southeast, i.e. towards the open part of the lake

3) A generally lower trophic state in Albania than in Montenegro

- According to the phytoplankton results from the two sampling campaigns, the Montenegrin part of Skadar Lake can be divided into 4 water bodies:

  1) Vucko mud (northwestern, largely isolated part of the lake; sampling point Kamenik)

  2) North coast (Plavnica and Podhum)

  3) South coast (Virpazar and Starcevo)

  4) Pelagial

- Results for the Albanian part of the lake were very similar and gave no reason to subdivide it further into separate water bodies.
- Results for the Albanian part of the lake were very similar and gave no reason to subdivide it further into separate water bodies.

6. Recommendations

The Water Framework Directive (WFD, 2000/60/EC) creates a new legislative framework to manage, use, protect, and restore surface and ground water resources within the river basins (or catchment areas) and in the transitional (lagoons and estuaries) and coastal waters in the European Union (EU). The WFD aims to achieve the sustainable management of water resources, to reach good ecological status, to prevent further deterioration of surface- and ground waters and to ensure sustainable functioning of aquatic ecosystems (as well as dependent wetlands and terrestrial systems).

The monitoring requirements according to Annex V of the WFD allow flexibility for the design of monitoring programs. Three different types of monitoring strategies are described: surveillance, operational and investigative monitoring, which all have different aims in terms of detecting or mapping of ecological status. Surveillance monitoring is carried out to assess the overall status of water bodies. Operational monitoring is employed for water bodies at risk of failing environmental objectives set according to the WFD, and to monitor the effect of measures devised to improve ecological status. Investigative monitoring serves to identify as yet unknown pressures.

The Water Framework Directive defines the status of water bodies by the extent of anthropogenically derived deviation from reference conditions, i.e. conditions that should occur at sites of any particular type in the absence of human impact. It is required that the ecological quality of water bodies should be classified into five quality classes (high, good, moderate, poor, and bad) using Ecological Quality Ratio (EQR), defined as the ratio between reference values (=no or little human impact) and observed values of the relevant biological quality element. However, the establishment of *reference conditions* of ecological status is very difficult for water bodies such as Skadar Lake for which few historical monitoring data exist. Besides, natural variability (that in principle are part of the reference conditions) may be higher than the variability caused by anthropogenic factors (because of the unique morphometry and hydrology of the lake, i.e. large and shallow polymictic lake with short residence time). The times at which monitoring is undertaken shall be selected so as to minimize the impact of seasonal variation, and thus to ensure that the results reflect changes in the status of water bodies as a result of changes due to anthropogenic pressure. Additional monitoring during different seasons may be carried out to achieve this objective for water bodies with high natural variability.

The following natural factors may have remarkable influence on the phytoplankton of Skadar Lake, causing deviations in seasonality and phenology: (1) Diurnal changes in physical, chemical and biological variables, some of which are regular due to daily cycles (e.g. photosynthesis and respiration, regular winds etc.), others being stochastic in nature (e.g. weather events). (2) Seasonal changes taking place at well-known regularity from year to year.

The sensitivity of lakes to natural variability factors depends strongly on their morphometry, and the role of physical drivers like wind, controlling the ecosystem processes increases with increasing lake area and decreasing depth (Nõges et al, 2009). Large and shallow polymictic lakes (like Skadar Lake) are extremely sensitive to natural physical drivers (Scheffer, 2004; Nõges et al., 2007). In this case, natural variability of phytoplankton community (that in principle should belong to reference conditions) exceeds often the variability caused by anthropogenic factors. Suitable aggregation of identified phytoplankton species into functional groups (Reynolds et al, 2002) might improve the predictability. For that reason, in future monitoring, more attention should be given to dominant *functional groups* of phytoplankton having in mind that these better represent anthropogenic impact.

High natural spatial and temporal variability of planktonic communities of Skadar Lake requires frequent sampling to ensure meaningful data for classification or detection of events (blooms). Although for phytoplankton parameters sampling is required to be carried out at least every 6 month (WFD Annex V, 1.3.4.), sampling frequency should be determined taking into consideration the natural variability of phytoplankton communities. Considering that Skadar Lake is a large and shallow polymictic lake, with high temporal variability of many physico-chemical parameters, a *monthly phytoplankton sampling* is recommended (during warm period of year at least) to minimize the impact of natural seasonal variation on the results.

Current phytoplankton community structure is a biological response to previous environmental conditions, with the time lag of the relationship determined by the time-scale over which phytoplankton gather resources and replicate. For this reason, the relationship between phytoplankton parameters and environmental parameters must be established. Therefore, phytoplankton sampling should be performed *together* with measurements of physico-chemical parameters.

When designing a monitoring program it is also important to consider spatial variations. Phytoplankton communities show marked spatial heterogeneity within large lakes as a result of patterns in lake circulation and mixing, differences in hydro-morphology and spatial gradients in flushing, grazing and nutrient availability.

According to the results of the present phytoplankton study, the Montenegrin part of Skadar Lake can be divided into four water bodies as described in chapter 5.

This means for future monitoring, it could be appropriate to define only four sampling points representing these water bodies.

Of all littoral sampling sites in Montenegro, Podhum has the least anthropogenic impact and its phytoplankton assemblage was most similar to the one in the pelagial zone and to those at the Albanian part of Skadar Lake.

1) "Vucko mud" water body. Kamenik is a sampling point located in Vucko mud – part of the lake with very limited hydrologic communication with the main water mass of the lake, especially after the construction of the Vranjina bridge. This site is under influence of Crnojevica River. Anthropogenic impacts comprise wastewater from a fish processing plant at Crnojevica River settlement, pollution from communal water from the city Cetinje via underground water and landfill leachate from the city of Cetinje. This part is also receives water from the right arm of Moraca river, which is the main source of water and nutrients of Skadar Lake. Phytoplankton analyses showed the highest trophic state at this sampling point. Therefore, Kamenik is representative sampling site for future monitoring of the Vucko mud water body.

2) “South coast” water body. The Starcevo sampling point is located at the south coast of Skadar Lake, though it has more pelagic then littoral characteristics owing to the steep shoreline. This site is located at the extension of the Moraca flow, which is especially visible after torrential rains. This corridor has specific hydrological and ecological characteristics. There is little local pollution but pollution from Moraca river and to some extent from the north coast of the lake. Therefore, Starcevo should be retained as a sampling point representative of the southern coast in future monitoring programs.

3) “North coast” water body. Plavnica is a sampling point located at the north coast of Skadar Lake, downstream from the mouth of Plavnicka river. It belongs to the littoral zone and has the lowest depth of all sampling points. Contamination and nutrient inputs from the city of Podgorica and agricultural activities in Zeta valley are evident (especially flushing of fertilizers during high water level). There is also local influence from transport and catering facilities in Plavnica settlement (restaurants, hotels, and marina). The northern coast of Skadar Lake gradually descends into the water and for that reason, this part of the lake is the shallowest, with extensive macrophyte cover and typical wetland characteristics. During high water level (winter), it is broadly flooded. During summer, a large part of the coast falls dry and turns into marshland. Plavnica is therefore a good representative of the northern coast of Skadar Lake and should be retained as a sampling point in future monitoring programs.

4) “Pelagial” water body. The center of the lake is the only typical pelagic point representing the ecological quality of the larger water mass of the lake and has the highest depth of all sampling points. For this reason, phytoplankton communities at this part of the lake are mostly represented by typical euplanktonic species, although littoral species are also present. The center of the lake might be considered as a reference point reflecting somewhat undisturbed conditions.

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8. Annexes: Summary tables of main results

Annex 1. Summary of the results of phytoplankton investigations at Lake Shkodra (Montenegro) in April and August 2013

		Sampling sites											
		Kamenik		Virpazar		Plavnica		Podhum		Pelagial		Starcevo	
Parameter	**Month**	**April**	**Aug.**	**April**	**Aug.**	**April**	**Aug.**	**April**	**Aug.**	**April**	**Aug.**	**April**	**Aug.**
Species number		37	41	29	39	26	35	22	40	19	20	42	31
Shannon diversity		2.33	3.99	2.10	3.94	2.23	3.64	1.48	4.49	2.91	2.13	2,8	3.01
Pielou's evenness		0.45	0.75	0.42	0.74	0.47	0.74	0.34	0.86	0.68	0.50	0.52	0.63
Total abundance (cells/L x 10^5)		70	40	66	32	44	18	20	5.5	6.5	35	35	39
Dominant species (% of total abundance)		FC 65.4	PL 27.4	FC 58.3	FU 24.9	DB 80.3	CG 27.3	DB 89.7	CG 21.7	DB 43.1	AI 56.3	FC 44.6	AI 39.7
Divisions (% of total abundance)													
– **Cyanophyta**		0.0	12.7	0.0	6.5	0.0	27.9	0.0	3.2	0.0	1.6	0.0	1.8
– **Bacillariophyta**		89.1	37.6	84.3	49.0	15.6	39.9	9.4	55.8	46.8	94.5	55.0	88.1
– **Chlorophyta**		1.8	41.9	1.5	29.4	4.0	12.9	0.6	37.7	4.7	3.8	6.3	10.1
– **Chrysophyta**		9.1	0.2	14.1	1.1	80.3	0.5	89.7	1.7	48.4	0.0	38.5	0.0
– **Pyrrophyta**		0.1	4.6	0.1	13.7	0.1	9.0	0.2	1.5	0.2	0.0	0.1	0.0
– **Euglenophyta**		0.0	3.0	0.0	0.3	0.0	9.9	0.0	0.2	0.0	0.0	0.0	0.0
Chlorophyll a concentration (µg/L)		6.2	12.5	5.3	8.7	4.1	6.9	7.7	2.8	2.8	2.1	8.0	3.3
TSI (based on chlorophyll a concentration)		48	56	47	51	44	49	50	37	40	36	51	43
Secchi disk (SD) transparency (m)		3.8	2.7	2.8	2.3	3.6	2.0	5.5	3.0	6.1	2.4	3.7	2.0
TSI (based on SD transparency)		67	71	72	74	68	78	62	70	60	72	68	78

Blue – oligotrophy, yellow – mesotrophy, green – eutrophy

FC = *Fragilaria crotonensis*; PL = *Planktonema lauterbornii*; FU = *Fragilaria ulna* var. *acus*;
DB = *Dinobryon bavaricum*; CG = *Cyclotella glomerata*; AI = *Aulacoseira italica*

Annex 2. Summary of the results of phytoplankton investigations at Lake Shkodra (Albania) in April and August 2013

		Sampling sites					
		Sterbeq		Zogaj		Shiroke	
Parameter	Month	April	Aug.	April	Aug.	April	Aug.
Species number		26	66	37	78	35	71
Shannon diversity		2.06	2.63	2.61	3.39	2.38	2.70
Pielou's evenness		0.54	0.58	0.68	0.75	0.62	0.60
Total abundance (cells/L x 10^5)		0.41	3.1	0.56	3.2	0.53	3.4
Dominant species (% of total abundance)		DB 58	CO 46	DB 47	CO 47	DB 39	CO 50
Divisions (% of total abundance)							
– Cyanophyta		0.0	18.7	0.0	21.8	0.0	20.5
– Bacillariophyta		22.4	48.8	32.2	49.3	26.3	52.1
– Chlorophyta		0.8	22.2	1.1	20.3	1.2	17.5
– Chrysophyta		69.6	2.9	49.4	2.6	52.4	2.3
– Pyrrophyta		7.2	7.4	17.3	6.0	20.1	7.6
– Euglenophyta		0.0	0.0	0.0	0.0	0.0	0.0
Chlorophyll a concentration (µg/L)		1.1	3.1	1.7	2.9	1.3	3.4
TSI (based on chlorophyll a concentration)		31	42	36	41	33	43
Secchi disk (SD) transparency (m)		2.75	2.3	3.62	2.6	2.87	2.1
TSI (based on SD transparency)		45	48	41	46	47	49

Blue – oligotrophy, yellow – mesotrophy

DB = *Dinobryon bavaricum*; CG = *Cyclotella ocellata*

Transboundary Monitoring Program

MACROPHYTES OF SKADAR LAKE

Final Report for Montenegro

Prepared by Sead Hadžiablahović

Environmental Protection Agency of Montenegro

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Contents

1. Introduction

The assessment of the structure and functioning of aquatic ecosystems, expressed as the deviation of the existing status from undisturbed (reference) conditions, is a requirement set forth by the European Directive 2000/60/EC, Water Framework Directive (WFD).

According to the WFD, macrophytes are one of several biological quality elements (BQE) to be used in ecological quality assessments of lakes (together with phytoplankton, macroinvertebrates and fish). As parameters to be studied, the WFD (Annex V) proposes:

1. Macrophyte composition
2. Total abundance
3. Average biomass

Aquatic vascular macrophytes, large water plants, visible to the naked eye, are widely utilized as reliable indicators of human disturbances in aquatic environments (Katnić 2007). Aquatic macrophytes in the littoral zones of lakes have two fundamental properties making them attractive as limnological indicators. Firstly, they react slowly and progressively to changes in nutrient conditions (Melzer 1999). Macrophytes therefore reflect in an integrated manner the environmental conditions to which they are subjected over longer periods of time and thus can be used as long-term indicators with high spatial resolution (Melzer 1999). They reflect patterns of nutrition or pollutant loads, caused by natural or anthropogenic inflows as well as by diffuse, non-point sources (Melzer 1999).

Other important environmental factors in lakes also have differentiating effects on macrophytes, e.g. light (Dale, 1986; Vant *et al.*, 1986; Tremp, 2007); sediment characteristics (Schneider and Melzer 2004; Paal *et al.,* 2007), trophic status (Schorer *et al.,* 2000; Kocic *et al.,* 2008) and hydrology (Tremolieres *et al.,* 1994; Madsen *et al.,* 2001).

The development of criteria and standards for evaluating the presence/ absence or extent of degradation of macrophyte vegetation makes it possible not only to take prevention and mitigation measures but also to conduct inventories and to design restoration measures (Armitage et al. 2002).

Skadar Lake

Lake Skadar is the largest lake on the Balkan Peninsula, located at the border between Montenegro and Albania. It is a transboundary Ramsar wetland site (No. 784 in 1995), and the Montenegrin part is a National Park since 1983. The surface area fluctuates seasonally between 370 and 600 km^2. The lake is not stratified owing to its shallow depth of 5 m (mean) to 8 m (maximum). The direct drainage basin covers 5,490 km^2, of which 4,460 km^2 are in Montenegro and 1,030 km^2 in Albania. The largest inflow is the Morača river (Montenegro), providing more than 62 % of the lake water, while Bojana river (Albania and Montenegro) flows out from the south end and drains into the Adriatic Sea. Lake Skadar has a subtropical climate and lies in the Submediterranean climate zone, resulting in high evaporation rates. The lake is borded to the southwest by high mountain ranges and rocky shorelines while it extends into a wide swamp area to the northeast providing an extensive semi-littoral zone with dense macrophyte cover. Frequent winds and the shallow depths prevent the formation of permanent thermal stratification.

2. Materials and methods

A preliminary field survey was done at the beginning of July 2013 to inspect suitable sampling sites that had been selected previously for the analysis of chemical parameters on the basis of known ecological disturbances of the lake according to data from the literature and information from institutions and researchers.

Five sampling stations on the Montenegrin side of Skadar Lake are designed as a system of five transects: NW- SE and SW-NE (Table 1)

Table 1. Sampling stations at Skadar Lake (Montenegrin side)

No.	Sampling station	Human impact/nature characteristics for water bodies	Coordinates
1.	Virpazar	Inflow of Virpazar canal	N 42^0 14 372 E 019^0 06 340
2.	Kamenik	Inflow of Crnojevića Rijeka	N 42^0 17 156 E 019^0 06 016
3.	Plavnica	Discharge of waste water and influence of ground waters	N 42^0 15 984 E 019^0 12 123
4.	Starčevo	Dilution of pollutants distant from the source of the pollution	N 42^0 11 082 E 019^0 12 150
5.	Podhum	Influence of KAP, surface waste waters and ground waters	N 42^0 16 462 E 019^0 21 178

At each sampling station the field protocol for surveying aquatic macrophytes was followed (General information, macrophyte survey - spatial structure of vegetation within transect, bottom quality, species composition on belt transect).

The lake bottom of the stations on the west side of the lake (Kamenik and Starčevo) is characterized by rocks, boulders and stones and differs from the one on the east side (Virpazar, Plavnica and Podhum) which is covered by sands.

Fig. 1. Sampling stations on the Montenegrin side of Skadar Lake

Methods for macrophyte sampling and assessment of trophic state of Skadar Lake

The sampling protocol for macrophytes used in the present study was as follows:

- Sample macrophytes according to the WISER method from the shoreline to the lower vegetation limit, during the period of maximum growth (mid-summer - 20th of July to 15th of August 2013).
- Determine specimens in the field and the laboratory using different floras and keys for vascular macrophytes (Tutin *et al.* 1968-1980, 1993).
- Prepare species lists for all stations (qualitative composition of macrophytes).
- Estimate the abundance of plant species present (quantitative composition of macrophytes) according to the five-point scale of Melzer (1999), and analyze the species according to the macrophyte indicator classification system of Melzer & Schneider (2001) (Table 2).
- Assess the trophic state of the of the water body based on the macrophyte vegetation.

Methodology of field survey

Transect method

The most widely applied method for aquatic vegetation surveys in both lakes and rivers is the transect method. The transect method is recommended by the European Committee for Standardization CEN (Comité Europeén de Normalisation) [CEN 2002, 2003]. The method consists in establishing transects (sectors) perpendicular to the lake shoreline, with a length covering the complete depth range of macrophyte growth, and in estimating the abundance and maximum colonization depth of each species identified within the transects.

WISER Method

The WISER method for lake macrophyte surveys is a modification of the transect method, widely used in European countries and compliant with the CEN standard, although CEN gives only broad guidance, leaving much space for interpretation.

Vegetation surveys along transects

1. A transect is a belt perpendicular to the shoreline extending from the shore to the point of maximum depth of plant growth and covering approximately two boat widths.
2. The starting points were situated at the beginning of the supra-littoral vegetation (wetland) or at the shoreline if easily recognizable.

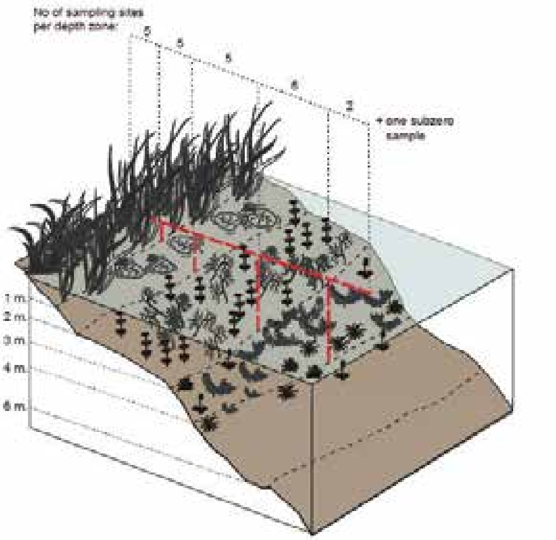

Fig. 2. Phytolittoral division of belt transects into depth zones and location of sampling sites according to the WISER method

3. Transects were divided into 1 m depth zones (1 m depth intervals). Within each depth zone, five evenly distributed sampling sites were determined (according to the pattern presented in Fig. 2).
4. To assure that the maximum depth of plant growth was defined properly, one sub-zero zone (i.e., no macrophyte present) was examined at the end of each transect.
5. Following the transect, two macrophyte samples were taken at each sampling site within a given depth zone, i.e. one sample from each side of the boat (Fig. 3).
6. At each sampling site within a depth zone, two macrophyte samples were taken, i.e. one sample from each side of the boat (Fig. 4).
7. The water-depth for each sample was recorded.
8. Based on two samples within a sampling site all species were identified and their abundance estimated in percentage continuous scale (easy to be recalculated to various point scales).
9. Overall abundance of the macrophyte cover within a sampling site was also determined.

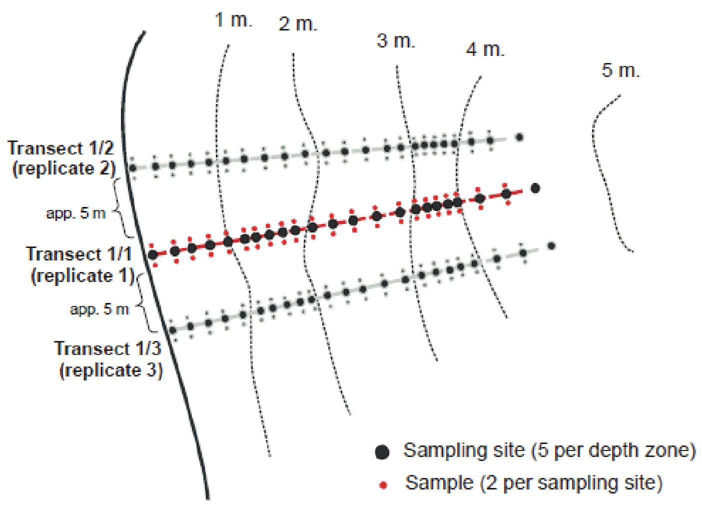

Fig. 3. Sampling pattern at sampling location (Transect three replicates)

Species identification and voucher specimens

- During the survey, all vascular plants were identified including floating leaved and submerged species.
- All specimens were determined to the species level.
- Voucher specimens of interesting species were deposited at the Herbarium of EPA.

Assessment of macrophyte abundance

Macrophyte abundance assessment methods vary considerably.
In the present study, abundance was estimated according to 5 categories: 1=very rare, 2=infrequent, 3=common, 4=frequent and 5=abundant or predominant (Melzer, 1999).

3. Data analysis and calculation of Macrophyte Index

All data collected at each sampling point, qualitative and quantitative, are used to calculate the Macrophyte Index (MI) according to the formula described by Melzer (1999):

$$MI = \frac{\sum_{i=1}^{n} I_i \cdot Q_i}{\sum_{i=1}^{n} Q_i}$$

where:

MI = Macrophyte Index
Ii = Indicator value of i-th species
Qi = Plant quantity of i-th species
n = total number of species with an indicator value

To calculate the Macrophyte Index, each abundance value was cubed (y = x3) because the correlation between the abundance estimation and plant quantity is not linear (Melzer, 1988; Kohler and Janauer, 1997) (Table 1.)

Table 1. Relationship between abundance scores and quantity

Five-degree-scale	Frequency	Plant quantity Q
1	very rare	1
2	infrequent	8
3	common	27
4	frequent	64
5	abundant	125

A classification scheme of nine indicator groups of macrophyte species was used to calculate the Macrophyte Index (Melzer and Schneider, 2001), ranging from 1 to 5 reflecting different sensitivities towards high nutrient loads (Table 2). Species belonging to indicator group 1 are restricted to oligotrophic conditions whereas those in indicator group 5 occur mainly in eutrophic, nutrient-rich lakes or sections of lakes. The remaining seven groups ranging from 1.5 to 4.5 represent transitions between these two extremes. Macrophyte species are assigned to certain indicator groups based on expert opinion and literature data. The mean MI of a lake is correlated with the total phosphorus concentration during the circulation time.

Table 2. Macrophyte indicator groups (Melzer and Schneider, 2001)

Indicator group 1.0	Indicator group 1.5	Indicator group 2.0
Chara hispida *Chara polyacantha* *Chara strigosa* *Potamogeton coloratus* *Utricularia stygia*	*Chara aspera* *Chara intermedia* *Utricularia minor*	*Chara delicatula* *Chara tomentosa* *Potamogeton alpinus*
Indicator group 2.5	**Indicator group 3.0**	**Indicator group 3.5**
Chara contraria *Chara fragilis* *Nitella opaca* *Nitellopsis obtusa* *Potamogeton gramineus* *Potamogeton natans* *Potamogeton x zixii*	*Chara vulgaris* *Myriophyllum spicatum* *Potamogeton filiformis* *Potamogeton perfoliatus* *Utricularia australis*	*Myriophyllum verticilatum* *Potamogeton berchtoldii* *Potamogeton lucens* *Potamogeton praelongus* *Potamogeton pusillus*
Indicator group 4.0	**Indicator group 4.5**	**Indicator group 5.0**
Hippuris vulgaris *Lagarosiphon major* *Potamogeton pectinatus*	*Elodea canadensis* *Elodea nuttallii* *Potamogeton compressus* *Potamogeton crispus* *Potamogeton obtusifolius* *Ranunculus circinatus* *Ranunculus trichophyllus*	*Ceratophyllum demersum* *Lemna minor* *Potamogeton mucronatus* *Potamogeton nodosus* *Sagittaria sagittifolia* *Spirodela polyrhiza* *Zannichelia palustris*

Six classes of the MI were designated, each representing different degrees of nutrient load. These six classes are assigned different colours (Table 3) to allow a clear illustration of the results.

Table 3. Relationship between index class, degree of nutrient status and assigned color

MI class	Degree of nutrient enrichment	
1.00–1.99	Slight	Dark blue
2.00–2.49	Low	Pale blue
2.50–2.99	Moderate	Green
3.00–3.49	Immense	Yellow
3.50–3.99	Heavy	Orange
4.00–5.00	Massive	Red

In the long term, it is foreseen to present results the software package ENSIS (Environmental Surveillance and Information System).

Table 4. Example: calculation of MI in Virpazar (depth zone 0-1 m and 1-2 m), the abundance values are cubed

Depth zone	Species list: Sampling sites within the depth zone	Transect 1/1					Transect 1/2					Transect 1/3					I	Q	$\sum I_i \times Q_i$
		1	2	3	4	5	1	2	3	4	5	1	2	3	4	5			
0-1 m	Ceratophyllum demersum	-	-	-	1	1	-	-	-	-	1	-	-	-	1	1	**5.0**	**5.0**	**25**
																			25
																			5

Depth zone	Species	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	I	Q	$\sum I_i \times Q_i$
1-2 m	*Ceratophyllum demersum*	8	8	27	8	-	-	8	27	8	-	-	8	27	8	-	5.0	137	685.0
	Potamogeton lucens	-	-	-	27	-	-	-	-	27	-	-	-	-	27	-	3.5	81	283.5
		$\sum Q_i$															218		968.5
																			4.44

4. Analysis and evaluation

Composition of macrophyte vegetation of Skadar Lake

Table 5. List of macrophyte plants sampled at Skadar Lake along five transects in summer 2013 (July – August). Numbers in bold signify indicator species according to Melzer and Schneider (2001).

No.	Scientific name	Family	Sampling points				
			1	2	3	4	5
1	*Butomus umbellatus*	Butomaceae	x				
2	*Calitriche hamulata*	Calitrichaceae					x
3	*Carex elata*	Cyperaceae		x			
4	*Carex* sp.	Cyperaceae			x		
5	*Carex* sp.	Cyperaceae					x
6	*Ceratophyllum demersum*	Ceratophyllaceae	x	x	x	x	x
7	*Cyperus longus*	Cyperaceae	x				
8	*Eleocharis acicularis*	Cyperaceae	x				
9	*Eleocharis palustris*	Cyperaceae	x				
10	*Fontinalis antipyretica*	Fontinalaceae		x			
11	*Galium palustre*	Rubiaceae	x				
12	*Gratiola officinalis*	Scrophulariaceae	x				
13	*Hydrocharis morsus ranae*	Hydrocharitaceae			x		
14	*Lysimachia vulgaris*	Myrsinaceae	x		x		
15	*Mentha aquatica*	Lamiaceae	x				x
16	*Myriophyllum spicatum*	Haloragaceae		x			
17	*Najas marina*	Najadaceae			x	x	x
18	*Najas minor*	Najadaceae			x		
19	*Nuphar lutea*	Nymphaeaceae	x	x	x		x
20	*Nymphaea alba*	Nymphaeaceae	x	x	x		x
21	*Oenanthe aquatica*	Umbeliferae	x				
22	*Phragmites australis*	Poaceae	x				x
23	*Polygonum amphibium*	Polygonaceae	x		x		
24	*Potamogeton crispus*	Potamogetonaceae			x		
25	*Potamogeton lucens*	Potamogetonaceae	x		x		x
26	*Potamogeton perfoliatus*	Potamogetonaceae					x
27	*Roripa amphibia*	Brassicaceae			x		x
28	*Salix alba*	Salecaceae			x		x
29	*Scirpus maritimus*	Cyperaceae	x	x	x		x
30	*Sparganium erectum*	Sparganiaceae	x				
31	*Spirodela polyrhyza*	Lemnaceae		x	x		
32	*Trapa natans*	Trapaceae	x	x	x	x	x
33	*Utricularia vulgaris*	Utriculariaceae	x	x	x		x
34	*Vallisneria spiralis*	Hydrocharitaceae		x	x	x	x

The total number of macrophyte species (shoreline, wetland and aquatic macrophytes) recorded at five selected sampling sites was as follows: Virpazar 19 species, Kamenik 11 species, Plavnica 18 species, Starčevo 4 species, and Podhum 16 species. Only six – mainly submerged living – of the altogether 34 species are assigned indicator values (*Ceratophyllum demersum*, *Myriophyllum spicatum*, *Potamogeton crispus*, *Potamogeton lucens*, *Potamogeton perfoliatus*, *Spirodela polyrhyza*). However, these species often occurred at high frequency and abundance. The maximum depth of plant growth recorded at both station was 4.3 m.

MI (Macrophyte Index) calculation

The MI of Skadar Lake in summer 2013 revealed eutrophic conditions. The southwestern and north-eastern shorelines of Lake Skadar were investigated at five localities.

The MI was calculated separately for each transect and depth zone. The results were as follows:

1. Virpazar - MI average all depths = **4.72**

MI according to the 4 different depth zones
Depth zone 0-1 m MI = 5.0
Depth zone 1-2 m MI = 4.44

2. Kamenik - MI average all depths = **4.98**

MI according to the 4 different depth zones
Depth zone 0-1 m MI = 4.97
Depth zone 1-2 m MI = 5.0
Depth zone 2-4 m MI = 4.98

3. Plavnica - MI average all depths = **4.99**

MI according to the 4 different depth zones
Depth zone 0-1 m MI = 4.96
Depth zone 1-2 m MI = 5.0
Depth zone 2-4 m MI = 5.0

4. Starčevo - MI average all depths = **5.0**

MI according to the 4 different depth zones
Depth zone 0-1 m MI = 5.0
Depth zone 1-2 m MI = 5.0
Depth zone 2-4 m MI = 5.0
Depth zone 4-6 m MI = 5.0

5. Podhum - MI average all depths = **3.5**

MI according to the 4 different depth zones
Depth zone 0-1 m MI = 3.51
Depth zone 1-2 m MI = 3.50
Depth zone 2-4 m MI = 3.50

The Macrophyte Index varied from 3.5 (at Podhum indicating heavy nutrient load) to 5.0 at Starčevo, thre latter indicating a massive nutrient load. The respective MI for the other stations indicated massive nutrient loads as well (Virpazar = 4.72; Kamenik = 4.98; Plavnica = 4.99; Starčevo = 5.0).

Species of indicator group 5.0 (*Ceratophyllum demersum*) and 3.5 (*Potamogeton lucens*) occurred frequently.

Ceratophyllum demersum had its highest abundance at Starčevo (west shore) while *Potamogeton lucens* was most abundant at Podhum.

The mean MI of the Skadar Lake (all stations together) is 4.64, idicating an overall massive nutrient load.

The Macrophyte Index of Skadar Lake hence indicates the trophic state (eutrophic) as well as the degree of nutrient load (massive).

The area in the vicinity of the two sampling sites at the eastern part of the lake (Plavnica and Podhum) as well as the sampling site at the western part (Virpazar) are impacted by intensive human activities - intensive agriculture and urbanization.

Also, human activities such as urban effluents and the lack of treatment of waste waters polluted by PCBs and heavy metals (red mud disposed by the KAP aluminium plant) endanger the stability of the Lake Skadar ecosystem.

Table 6. Summary of data analyses and the degree of nutrient enrichment for all sampling points at Lake Skadar (Montenegrin side, summer 2013)

Lake Skadar	Sampling point	No. of species recorded	No. of species with indicator value	Max. depth of plants growth	MI	Degree of nutrient enrichment
	MNE 1	19	2	1.9 m	4.72	Massive
	MNE 2	11	3	3.2 m	4.98	Massive
	MNE 3	18	4	3.9 m	4.99	Massive
	MNE 4	4	2	4.2 m	5.00	Massive
	MNE 5	16	3	3.2 m	3.50	Heavy
All stations	1+2+3+4+5	34	7		4.64	Massive

5. Conclusions and recommendations

In July/August 2013 a macrophyte survey was conducted at Skadar Lake in the frame of the "Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar (CSBL)" Project.

The aim of the survey was to estimate the ecological status of Skadar Lake using macrophytes, corresponding to the requirements of the European Water Framework Directive (WFD).

The total number of plant species recorded during the investigation of macrophytes at five selected stations was 34 (Virpazar 19 species, Kamenik 11 species, Plavnica 18 species, Starčevo 4 species, Podhum 16 species). Indicator values are assigned to only six of these species, meaning that the vast majority of the species were not included in the MI calculation. However, the identified indicator species often occurred at high frequency and/or abundance.

According to the proposed methodology (Melzer 1999), the Macrophyte Index of Skadar Lake shows that the status of the lake as a whole **is not** satisfactory even though the lake has a great self-purification capacity due to its short residence time (The lake water changes 3 to 4 times per year).

The results indicated that Skadar Lake is impacted by human activities. The nutrient enrichment was massive at four and heavy at one sampling site. MI indicates heavy nutrient enrichment at Podhum and massive nutrient enrichment at Virpazar, Kamenik, Plavnica and Starčevo.

However, these data cannot indicate the ecological state of whole lake, because the number of transects should be much higher. The recommended number of transects in relation for lakes with a surface area of more than 100 km^2, like Skadar Lake, the number of transects should be 30-50 (Schaumburg et al. 2007; Kashta, 2013).

The Macrophyte Index of Melzer (1999), which was developed for water bodies in temperate zones of Central Europe, was applied for the first time to Skadar Lake. The extent to which it is applicable to Mediterranean limnosystems remains to be seen. In the present study, between 11% and 29% of the plant species found at the five sampling sites are not listed as indicators by Melzer and Schneider (2001). This may have introduced a bias in the calculated MIs if these species are abundant. However, the percentage was much higher in deeper depth strata and when considering only submerged and free floating macrophyte species. More research is needed in this respect to establish the indicator values of macrophyte species of Mediterranean limnosystems. As a consequence of the regionally differing behaviour of some species, an overall application of the Macrophyte Index is not possible without a screening of the composition of characteristic macrophyte communities and a re-evaluation of the indicator groups defined by Melzer (1999).

Wastewater from urban and industrial centers in Montenegro (major towns Podgorica, Nikšić and Cetinje) is discharged into Lake Skadar with minimum or no treatment at all.

Also, human activities such as agriculture, illegal construction, urban effluents and the lack of treatment of waste waters polluted by PCBs and heavy metals (red mud disposed by the KAP aluminium plant), endanger the stability of the Lake Skadar ecosystem.

Dynamic water flow, which is prevalent at Lake Skadar due to variable substrate permeability and inflow of fresh cold water from mountain rivers, streams and underground waters could explain differences in MI between polluted and unpolluted sites.

Before drawing conclusions, it is necessary to relate the Macrophyte Indices established in the present study with result from chemical analyses conducted in parallel at the same stations.

The results of the present study can be used as baseline data. It would be necessary, in the future, to determine the Macrophyte Index annually. This would enable more profound and comprehensive assessments, monitoring and using aquatic plants as indicators of water quality, due to their conditional reaction on pollution, easy application and detection.

More people and funding should be made available to study this important limnosystem which is the largest in Balkan. These goals can be achieved among others by conducting comprehensive and longer-term studies and by improving institutional cooperation and data analysis sharing (Katnić 2007).

6. References

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Annexes

Annex 1. Field protocols

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 1/4
GENERAL INFORMATION				
Lake name: Skadar/Shkodra lake	**Lake type:**	**Lake code:**	**Surveyor:**	**Date:**
Lake information				
Surface area (km^2)	370 to 530 km	Lake altitude (m asl)		
Maximum depth (m)	44 m (144 ft)	Mean alkalinity (meq)		
Mean depth (m)		Water colour (mg/Pt/L)		
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)		C
Retention time		Dominating land use in cathcment (F=forest, A=agricultural, U=urban, O=other)		U
P of locality				
Locality number: 1	**Starting time:**	**Ending time:**		
Start-point (GPS)	N 42^0 14 372 E 019^0 06 340			
End-point (GPS)	N 42^0 14 821 E 019^0 06 580			
Locality direction (degrees)	SOUTHWEST - NORTHEAST			
Freehand drawing of transect				
VIRPAZAR Coast: Salix alba, Vitex agnus castus, Amorpha fruticosa, Perploca graeca, Paspalum paspalodes, Teucrium scorpioides, Alisma lanceolatum ...				

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE					Page 2/4
MACROPHYTE SURVEY					
Transect no:		**Transect .../1**	**Transect .../2**	**Transect .../3**	
Vegetation spatial structure within a transect					
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids					
Growth form dominating		NY	NY	NY	
Start- point depth (m)		0.05	0.05	0.05	
End-point depth (m)		2.0	2.0	2.0	
Maximum depth of plant growth (m)		1.9	1.9	1.9	
Max. observed depth of rush (m)					
Max. observed depth of elodeids (m)					
Max. observed depth of charids (m)		-	-	-	
Max. observed depth of isoetids (m)		-	-	-	
Bottom quality: *total cover of whole zone as percentage*					
Rock (>4000 mm)					
Boulders (250-4000 mm)					
Stone (16-250 mm)					
Gravel (2-16 mm)					
Sand (0.06-2 mm,)		+	+	+	
Silt (smooth between fingers)					
Clay (elastic, grey)					
Mud					
Peat					
Detritus (tree leaves, trash, etc.)		+	+	+	
Other (specify)					
Species composition on a belt transect (abundance/cover; 0.5,1 ,3 ,5 ,7 ,10 ,15, 20, 30...100%)					

Depth zone	Species list:	**Transect .../1**					**Transect .../2**					**Transect .../3**				
	Sampling sites within the depth zone:	**1 0.1**	**2 0.3**	**3 0.7**	**4 0.9**	**5 1.0**	**1 0.1**	**2 0.3**	**3 0.7**	**4 0.9**	**5 1.0**	**1 0.1**	**2 0.3**	**3 0.7**	**4 0.9**	**5 1.0**
0-1 m continued																
	Nymphaea alba		2	5	5	5			5	5	5			2	2	2
	Nuphar lutea		2	2	2	2			2	2	2			2	2	2
	Phragmites australis		5	2				2	2				3	2		
	Mentha aquatica	4					4					4				
	Lysimachia vulgaris	2					3					2				
	Galium palustre	1														
	Eleocharis palustris	2										3				
	Eleocharis acicularis	2														
	Sparganium erectum	2										2				
	Gratiola officinalis	3					3					3				
	Cyperus longus	3														
	Butomus umbelatus	2					2									
	Sparganium erectum	2					2									
	Oenanthe aquatica	1														
	Polygonum amphibium	1														
	Scirpus maritimus		2					3					2			
	Utricularia vulgaris				2					2						
	Ceratophyllum demersum	-	-	-	1	1	-	-	-	1	-	-	-	-	1	1

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 1.2	2 1.3	3 1.5	4 1.8	5 1.9	1 1.2	2 1.3	3 1.5	4 1.8	5 1.9	1 1.2	2 1.3	3 1.5	4 1.8	5 1.9
1-2 m																
	Nuphar lutea	5	5	2			5	5				5	5			
	Nymphaea alba	3	2	3			3		3			3		2		
	Trapa natans		2	4					4					4		
	Ceratophyllum demersum	-	2	2	3	2	-	-	2	3	2	-	-	2	3	2
	Potamogeton lucens	-	-	-	3	-	-	-	-	3	-	-	-	-	3	-

Virpazar - start point

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 1/4
GENERAL INFORMATION				
Lake name: Skadar/Shkodra lake	**Lake type:**	**Lake code:**	**Surveyor:**	**Date:**
Lake information				
Surface area (km²)	370 to 530 km	Lake altitude (m asl)		
Maximum depth (m)	44 m (144 ft)	Mean alkalinity (meq)		
Mean depth (m)		Water colour (mg/Pt/L)		
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)		C
Retention time		Dominating landuse in cathcment (F=forest, A=agricultural, U=urban, O=other)		O
P of locality				
Locality number: 2	**Starting time:**	**Ending time:**		
Start-point (GPS)	N 42⁰ 17 156 E 019⁰ 06 016			
End-point (GPS)	N 42⁰ 17 309 E 019⁰ 06 080			
Locality direction (degrees)	SOUTHWEST - NORTHEAST			
Freehand drawing of transect				
KAMENIK Coast: Vitex agnus castus, Salix alba, Perploca graeca, Cyperus longus, Scirpus maritimus, Carex elata, Lysimachia vulgaris, Alisma lanceolatum, Paspalum paspalodes				

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE	Page 2/4
MACROPHYTE SURVEY	

Transect no:		Transect .../1	Transect .../2	Transect .../3
Vegetation spatial structure within a transect				
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids				
Growth form dominating		NY	NY	NY
Start- point depth (m)		0.05	0.05	0.05
End-point depth (m)		3.2	3.2	3.2
Maximum depth of plant growth (m)		3.2	3.2	3.2
Max. observed depth of rush (m)				
Max. observed depth of elodeids (m)		3.2	3.2	3.2
Max. observed depth of charids (m)		-	-	-
Max. observed depth of isoetids (m)		-	-	-
Bottom quality: *total cover of whole zone as percentage*				
Rock (>4000 mm)		+	+	+
Boulders (250-4000 mm)		+	+	+
Stone (16-250 mm)				
Gravel (2-16 mm)				
Sand (0.06-2 mm,)		+	+	+
Silt (smooth between fingers)				
Clay (elastic, grey)				
Mud				
Peat				
Detritus (tree leaves, trash, etc.)				
Other (specify)				

Species composition on a belt transect (abundance/cover; 0.5,1 ,3 ,5 ,7 ,10 ,15, 20, 30...100%)

Depth zone	Species list: Sampling sites within the depth zone:	Transect .../1					Transect .../2					Transect .../3				
		1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
0-1 m continued (0-4 m from the shoreline)																
	Fontinalis antipyretica	3														
	Carex elata	2					2									
	Scirpus maritimus	3					3					3				
	Vallisneria spiralis	2					2					2				
	Trapa natans	2					2					2				
	Nuphar lutea	2					2					2				
	Ceratophyllum demersum	3					3					2				
	Nymphaea alba	4					4					4				
	Utricularia vulgaris	1														
	Myriophyllum spicatum	1														

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
1-2 m (4-10 m from the shoreline)																
	Nymphaea alba	4					4					4				
	Nuphar lutea											2				
	Trapa natans	2					2					2				
	Ceratophyllum demersum	3					3					3				
	Spirodela polyrhyza											1				

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 2.5	2 2.7	3 2.8	4 2.9	5 2.9	1 2.5	2 2.7	3 2.8	4 2.9	5 2.9	1 2.5	2 2.7	3 2.8	4 2.9	5 2.9
2-3 m																
	Trapa natans	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
	Ceratophyllum demersum	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
	Myriophyllum spicatum	1														

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 3.0	2 3.1	3 3.2	4 3.2	5 3.2	1 3.0	2 3.1	3 3.2	4 3.2	5 3.2	1 3.0	2 3.1	3 3.2	4 3.2	5 3.2
3-4 m																
	Ceratophyllum demersum	2	2	1	1	1	2	2	1	1	1	2	2	1	1	1

Kamenik - start point

<table>
<tr><td colspan="4">FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE</td><td rowspan="2">Page 1/4</td></tr>
<tr><td colspan="4">GENERAL INFORMATION</td></tr>
<tr><td>Lake name:
Skadar/Shkodra lake</td><td>Lake type:</td><td>Lake code:</td><td>Surveyor:</td><td>Date:</td></tr>
<tr><td colspan="5">Lake information</td></tr>
<tr><td>Surface area (km²)</td><td>370 to 530 km</td><td colspan="2">Lake altitude (m asl)</td><td></td></tr>
<tr><td>Maximum depth (m)</td><td>44 m (144 ft)</td><td colspan="2">Mean alkalinity (meq)</td><td></td></tr>
<tr><td>Mean depth (m)</td><td></td><td colspan="2">Water colour (mg/Pt/L)</td><td></td></tr>
<tr><td>Lake perimeter incl. islands (km)</td><td></td><td colspan="2">Dominating geology in catchment
(S=siliceous, C=calcareous, O=organic)</td><td>C</td></tr>
<tr><td>Retention time</td><td></td><td colspan="2">Dominating landuse in cathcment
(F=forest, A=agricultural, U=urban, O=other)</td><td>A</td></tr>
<tr><td colspan="5">P of locality</td></tr>
<tr><td>Locality number: 3</td><td colspan="2">Starting time:</td><td colspan="2">Ending time:</td></tr>
<tr><td>Start-point (GPS)</td><td colspan="2">N 42⁰ 15 984 E 019⁰ 12 123</td><td colspan="2"></td></tr>
<tr><td>End-point (GPS)</td><td colspan="2">N 42⁰ 14 882 E 019⁰ 11 539</td><td colspan="2"></td></tr>
<tr><td>Locality direction (degrees)</td><td colspan="4">NORTHEAST- SOUTHWEST</td></tr>
<tr><td colspan="5">Freehand drawing of transect</td></tr>
<tr><td colspan="5">PLAVNICA
Coast:
Salix alba, Nymphaea alba, Lysimachia vulgaris, Polygonum amphibium, Roripa amphibia, Carex sp.</td></tr>
</table>

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 2/4
MACROPHYTE SURVEY				
Transect no:		**Transect .../1**	**Transect .../2**	**Transect .../3**
Vegetation spatial structure within a transect				
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids				
Growth form dominating		NY	NY	NY
Start- point depth (m)		0.05	0.05	0.05
End-point depth (m)		3.9	3.9	3.9
Maximum depth of plant growth (m)		3.9	3.9	3.9
Max. observed depth of rush (m)				
Max. observed depth of elodeids (m)		3.9	3.9	3.9
Max. observed depth of charids (m)		-	-	-
Max. observed depth of isoetids (m)		-	-	-
Bottom quality: *total cover of whole zone as percentage*				
Rock (>4000 mm)				
Boulders (250-4000 mm)				
Stone (16-250 mm)				
Gravel (2-16 mm)				
Sand (0.06-2 mm,)		+	+	+
Silt (smooth between fingers)				
Clay (elastic, grey)				
Mud		+	+	+
Peat				
Detritus (tree leaves, trash, etc.)				
Other (specify)				

Species composition on a belt transect (abundance/cover; 0.5,1 ,3 ,5 ,7 ,10 ,15, 20, 30...100%)																
Depth zone	Species list:	**Transect .../1**					**Transect .../2**					**Transect .../3**				
	Sampling sites within the depth zone:	**1 .05**	**2 0.2**	**3 0.5**	**4 0.8**	**5 0.9**	**1 .05**	**2 0.2**	**3 0.5**	**4 0.8**	**5 0.9**	**1 .05**	**2 0.2**	**3 0.5**	**4 0.8**	**5 0.9**
0-1 m continued																
	Nymphaea alba	4	2	4	2	2	4	2	4			4	2	4	2	2
	Polygonum amphibium	4		2			4		2			4				
	Nuphar lutea		5	2	3	3		5	2	3	3		5	3	3	3
	Carex sp.	2														
	Lysimachia vulgaris	2					2					2				
	Roripa amphibia	2	2					2					2			
	Ceratophyllum demersum	2	2	2	3	3	2	2	2	3	3	2	2	2	3	3
	Utricularia vulgaris	2	2				2	2	2			2	2			
	Scirpus maritimus	2	2	2	3		2	2	2	3		2	2	2	3	
	Hydrocharis morsus ranae	1														
	Vallisneria spiralis		3	2	2	2		2	2	2	2		2	2	2	2
	Spirodela polyrhyza		1													
	Salix alba	1						1								
	Potamogeton crispus				1											
	Potamogeton lucens				2	2										
	Najas marina				2	2				2	2				2	2
	Najas minor				2	2										
	Trapa natans				2	2										

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 1.2	2 1.3	3 1.5	4 1.7	5 1.8	1 1.2	2 1.3	3 1.5	4 1.7	5 1.8	1 1.2	2 1.3	3 1.5	4 1.7	5 1.8
1-2 m continued																
	Ceratophyllum demersum	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
	Vallisneria spiralis	3	2	2			3	2	2			3	2	2		
	Najas marina						2									

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 2.2	2 2.4	3 2.5	4 2.7	5 2.9	1 2.2	2 2.4	3 2.5	4 2.7	5 2.9	1 2.2	2 2.4	3 2.5	4 2.7	5 2.9
2-3 m continued																
	Ceratophyllum demersum	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
	Vallisneria spiralis	3	2	2			3	2	2			3	2	2		
	Najas marina						2									

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 3.1	2 3.3 1.4	3 3.5	4 3.7	5 3.9	1 3.1	2 3.3 1.4	3 3.5	4 3.7	5 3.9	1 3.1	2 3.3 1.4	3 3.5	4 3.7	5 3.9
3-4 m continued																
	Ceratophyllum demersum	3	2	2	1	1	3	2	2	1	1	3	2	2	1	1

Plavnica - start point

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 1/4
GENERAL INFORMATION				
Lake name:	**Lake type:**	**Lake code:**	**Surveyor:**	**Date:**

Lake information			
Surface area (km^2)		Lake altitude (m asl)	
Maximum depth (m)		Mean alkalinity (meq)	
Mean depth (m)		Water colour (mg/Pt/L)	
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)	
Retention time		Dominating landuse in cathcment (F=forest, A=agricultural, U=urban, O=other)	

P of locality		
Locality number:	**Starting time:**	**Ending time:**
Start-point (GPS)	N 42^0 11 501 E 019^0 11 680	
End-point (GPS)	N 42^0 11 673 E 019^0 12 668	
Locality direction (degrees)	SOUTHWEST - NORTHEAST	

Freehand drawing of transect
STARČEVO Coast: near to the coast Vitex agnus castus, Salix alba, Fraxinus angustifolia, Cyperus longus

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE			Page 2/4
MACROPHYTE SURVEY			
Transect no:	**Transect .../1**	**Transect .../2**	**Transect .../3**
Vegetation spatial structure within a transect			
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids			
Growth form dominating	NY	NY	NY
Start- point depth (m)	0.1	0.1	0.1
End-point depth (m)	4.3	4.3	4.3
Maximum depth of plant growth (m)	4.2	4.0	4.0
Max. observed depth of rush (m)			
Max. observed depth of elodeids (m)	-	-	-
Max. observed depth of charids (m)	-	-	-
Max. observed depth of isoetids (m)	-	-	-
Bottom quality: *total cover of whole zone as percentage*			
Rock (>4000 mm)	+	+	+
Boulders (250-4000 mm)	+	+	+
Stone (16-250 mm)			
Gravel (2-16 mm)			
Sand (0.06-2 mm,)	+	+	+
Silt (smooth between fingers)			
Clay (elastic, grey)			
Mud			
Peat			
Detritus (tree leaves, trash, etc.)			
Other (specify)			

Species composition on a belt transect (abundance/cover; 0.5,1 ,3 ,5 ,7 ,10 ,15, 20, 30...100%)

Depth zone	Species list:	**Transect .../1**					**Transect .../2**					**Transect .../3**				
	Sampling sites within the depth zone:	**1 0.5-**	**2**	**3**	**4**	**5**	**1 0.5**	**2**	**3**	**4**	**5**	**1 0.5**	**2**	**3**	**4**	**5**
0-1 m continued (0-2,5 m from the shoreline)																
	Vallisneria spiralis	4					4					4				
	Ceratophyllum demersum	2					2					2				
	Najas marina	1					1					1				

Depth zone	Species list:	**Transect .../1**					**Transect .../2**					**Transect .../3**				
	Sampling sites within the depth zone:	**1 1.7**	**2**	**3**	**4**	**5**	**1 1.7**	**2**	**3**	**4**	**5**	**1 1.5**	**2**	**3**	**4**	**5**
1-2 m (2,5-6 m from the shoreline)																
	Vallisneria spiralis	3					3					3				
	Ceratophyllum demersum	3					3					3				
	Najas marina	1					1					3				
	Trapa natans	1					-					-				

Depth zone	Species list:	Transcect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 2.8	2	3	4	5	1 2.7	2	3	4	5	1 2.5	2	3	4	5
2-3 m (6-9 m from the shoreline)																
	Vallisneria spiralis	3					3					3				
	Ceratophyllum demersum	3					3					3				
	Najas marina	1					1					3				

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 3.2	2 3.4	3 3.6	4 3.7	5 3.9	1 3.2	2 3.4	3 3.6	4 3.7	5 3.9	1 3.2	2 3.4	3 3.6	4 3.7	5 3.9
3-4 m (200-500 m from the shoreline)																
	Vallisneria spiralis	2	3	4	4	4	2	3	3	4	4	2	2	3	4	4
	Ceratophyllum demer-sum	3	3	2	2	1	3	3	2	2	1	3	3	2	2	1
	Najas marina	3	2	1	1		3	2	1	1		3	2	1	1	

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 4.0	2 4.2	3 4.3	4	5	1 4.0	2 4.2	3 4.3	4	5	1 4.0	2 4.2	3 4.3	4	5
4-5 m (500-1000 m from the shoreline)																
	Vallisneria spiralis	2	1	-			-	-	-			-	-	-		
	Ceratophyllum demersum	2	1	-			2	-	-			2	-	-		

Starčevo - start point

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE GENERAL INFORMATION				Page 1/4
Lake name:	**Lake type:**	**Lake code:**	**Surveyor:**	**Date:**
Lake information				
Surface area (km^2)		Lake altitude (m asl)		
Maximum depth (m)		Mean alkalinity (meq)		
Mean depth (m)		Water colour (mg/Pt/L)		
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)		
Retention time		Dominating land use in cathcment (F=forest, A=agricultural, U=urban, O=other)		
P of locality				
Locality number:	**Starting time:**		**Ending time:**	
Start-point (GPS)	N 420 16 675 E 0190 21 658			
End-point (GPS)	N 420 16 198 E 0190 21 734			
Locality direction (degrees)	NORTHWEST - SOUTHEAST			
Freehand drawing of transect				
PODHUM Coast: Salix alba, Phragmites australis, Carex sp.				

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE MACROPHYTE SURVEY				Page 2/4
Transect no:		**Transect .../1**	**Transect .../2**	**Transect .../3**
Vegetation spatial structure within a transect				
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids				
Growth form dominating		NY	NY	NY
Start- point depth (m)		0.05	0.05	0.05
End-point depth (m)		3.4	3.4	3.4
Maximum depth of plant growth (m)				
Max. observed depth of rush (m)				
Max. observed depth of elodeids (m)				
Max. observed depth of charids (m)		-	-	-
Max. observed depth of isoetids (m)		-	-	-
Bottom quality: *total cover of whole zone as percentage*				
Rock (>4000 mm)				
Boulders (250-4000 mm)				
Stone (16-250 mm)				
Gravel (2-16 mm)				
Sand (0.06-2 mm,)		+	+	+
Silt (smooth between fingers)				
Clay (elastic, grey)				
Mud		+	+	+
Peat				
Detritus (tree leaves, trash, etc.)		+	+	+
Other (specify)				
Species composition on a belt transect (abundance/cover; 0.5,1 ,3 ,5 ,7 ,10 ,15, 20, 30...100%)				

Depth zone	Species list: Sampling sites within the depth zone:	Transect .../1					Transect .../2					Transect .../3				
		1 **0.0**	**2** **0.2**	**3** **0.5**	**4** **0.7**	**5** **0.9**	**1** **0.0**	**2** **0.2**	**3** **0.5**	**4** **0.7**	**5** **0.9**	**1** **0.0**	**2** **0.2**	**3** **0.5**	**4** **0.7**	**5** **0.9**
0-1 m continued																
	Salix alba	3					3					3				
	Phragmites australis	4					4					4				
	Carex sp.	2														
	Nuphar lutea		4	4	4	4		4	4	4	4		4	4	4	4
	Nymphaea alba		2	2	2	2		2	2	2	2		2	2	2	2
	Ceratophyllum demersum	-	-	-	-	1	-	-	-	-	1	-	-	-	-	-
	Utricularia vulgaris		1	2	3	1		1	2	3	1		1	2	3	1
	Scirpus maritimus		1	2	2	3		1	2	3	3		1	2	3	3
	Potamogeton lucens		2	3	2	2		2	3	2	2		2	3	2	2
	Potamogeton perfoliatus				1	1				1					1	
	Roripa amphibia		1		1											
	Mentha aquatica		1													
	Trapa natans		2			1										
	Vallisneria spiralis				1	1										
	Calitriche hamulata		1													
	Najas marina					1					1					

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	**1** **1.0**	**2** **1.2**	**3** **1.5**	**4** **1.8**	**5** **1.9**	**1** **1.2**	**2** **1.3**	**3** **1.5**	**4** **1.8**	**5** **1.9**	**1** **1.2**	**2** **1.3**	**3** **1.5**	**4** **1.8**	**5** **1.9**
1-2 m(300 to 500 m from the shoreline)																
	Nuphar lutea	4	3													
	Nymphaea alba	2	2													
	Potamogeton lucens	3	3	4	4	4	-	-	4	4	4	-	-	4	4	4
	Potamogeton perfoliatus	1														
	Scirpus maritimus	3					3									
	Phragmites australis													3		

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	**1** **2.2**	**2** **2.4**	**3** **2.5**	**4** **2.7**	**5** **2.9**	**1** **2.2**	**2** **2.4**	**3** **2.5**	**4** **2.7**	**5** **2.9**	**1** **2.2**	**2** **2.4**	**3** **2.5**	**4** **2.7**	**5** **2.9**
2-3 m(500 to 1200 m from the shoreline)																
	Potamogeton lucens	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
	Najas marina	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
	Najas minor			1												
	Vallisneria spiralis			1												

Depth zone	Species list:	Transect .../1					Transect .../2					Transect .../3				
	Sampling sites within the depth zone:	1 3.1	2 3.2	3 3.4	4	5	1 3.1	2 3.2	3 3.4	4	5	1 3.1	2 3.2	3 3.4	4	5
3-4 m (500 to 1200 m from the shoreline)																
	Potamogeton lucens	2	1	-			2	1	-			1	1	-		
	Najas marina	2	1	-			2	1	-			1	1	-		

Podhum - start point

Project

MACROPHYTES OF LAKES PRESPA AND SHKODRA

Trans-boundary Monitoring program

Prepared by

Lefter Kashta and Marash Rakaj

January 2013

Contents

1. Introduction

The assessment of the structure and functioning of an aquatic ecosystem, expressed as the deviation of the existing status from undisturbed (reference) conditions, is the requirement set for the monitoring systems by European Directive 2000/60/EC, called the Water Framework Directive [WFD; EC 2000].
The ecological status of an ecosystem is assessed based on the condition of the aquatic communities, the so-called biological quality elements (BQEs) (Kolada et al., 2012). The Water Framework Directive (WFD) has made the survey of macrophytes obligatory: they are one of four biological elements needed to assess the ecological status of surface waters.
Aquatic macrophytes in the littoral zones of lakes have two fundamental properties, which make them attractive as limnological indicators. Firstly, they react slowly and progressively to changes in nutrient conditions. Secondly, the littoral zone may experience patterns of nutrient (and pollutant) concentrations caused by natural or artificial inflows as well as by diffuse, non-point sources (Melzer, 1999).
According to the WFD-EU, species composition and abundance of species are the basic parameters for the assessment of the ecological status (Janauer, 2002).
The monitoring of all biological elements, including macrophytes, requires the development of two complementary methodologies: (i) the methodology of a field survey, i.e. the manner of material sampling; (ii) the methodology for water assessment based on the material collected in the field, i.e. the manner of data analysis and calculation of biological indicators (Kolada et al., 2012).
Development of criteria and standards by which to evaluate the presence/absence or extent of degradation not only would serve in prevention and mitigation but would greatly assist inventory and restoration activities (Armitage et al. 2002).
The aim of the survey is to evaluate the ecological status of lakes Shkodra and Prespa according to the Water Framework Directive (WFD) using macrophytes.

Objectives

- Sampling of macrophytes from the shoreline to the lower vegetation limit and preparing the list of species present at selected sampling sites (qualitative composition of macrophytes).
- Estimation of plant abundance of species present (quantitative composition of macrophytes) according to a five-point scale (Melzer 1999)
- Calculation of trophic state at each station according to the species recorded to which an indicator value has been assigned (Melzer and Schneider, 2001).

2. Parameter and sampling points

Three sampling stations on the Albanian side of Shkodra Lake and two stations on the Albanian side of Prespa Lake were selected for macrophyte sampling. These stations have been previously selected for sampling chemical parameters based on recognized zones of ecological disturbance in the lakes according to data from the literature (Table 1, Figure 1).

At each point, a survey of macrophyte species, along a bellt transect according to WISER method, was performed. All data collected at each sampling point, qualitative and quantitative, were used to calculate the Macrophyte Index (Melzer, 1999).

Shkodra Lake is the largest lake on the Balkan Peninsula in terms of water surface. The drainage area of the lake is about 5,500 km2 (4,470 km2 in Montenegro and 1,030 km2 in Albania). The lake area varies between 353 km2 in dry periods and 500 km2 in wet periods (Keukelaar et al., 2006).

Table 1: List of sampling points in Albanian sides of Shkodra and Prespa Lakes

Lake	Sampling point	Coordinates
Shkodra Lake	AL 1 Sterbeq	N 42^0 11’ 45.8“ E 19^0 23’ 21.8“
	AL 2 Zogaj	N 42^0 04’ 22.6“ E 19^0 24’ 13.9“
	AL 3 Shiroka	N 42^0 03’ 41.3” E 19^0 27’ 13.9“
Prespa Lake	AL 1 Gollomboc	N 40^0 51’ 43.8” E 20^0 56’ 25.4“
	AL 2 Pustec	N 40^0 47’ 27.2“ E 20^0 54’ 42.2“

The lake has a peculiar water regime, with water level fluctuations of up to five metres. The Moraca River, with its two tributaries, Zeta and Cijevna/Cemi, contributes 62 percent of the lake’s water. About 30 percent of it comes from underground springs called “eyes.” The rest comes directly from the mountains or from rainfall.

Its northern coast is flat, gradually descending toward the lake, and it is covered with lush vegetation. The southern coast is steep and rugged. Shkoder Lake is relatively shallow and the deepest part of the lake bed lies below sea level, meaning the lake lies in a crypto-depression (Marczin, 2007).

a b

Figure 1: Maps with sampling points on the Albanian sides of **a:** Shkodra and **b.** Prespa lakes

The Macro Prespa Lake, with a total surface ca. 259.4 km^2, is shared between three countries, FYR of Macedonia, Greece and Albania (Albanian part ca. 45.5 km^2). The lake lies at an altitude of 853m; it is 54 m deep, and it has low transparency (from 2m to 6m). The water level of Macro Prespa has decreased during recent years by approximately 8m, however, the causes of this phenomenon have not yet been fully investigated. Macro Prespa is classified as oligotrophic and has good water oxygenation, but its transparency is constantly decreasing and it seems to be increasingly burdened with pollutants (SPP-WWF, 2002).

Data provided

Qualitative: macrophyte species composition

Quantitative: macrophyte species abundance

Parameter calculated

MI (Macrophyte Index)

3. Methods for macrophytes sampling and assessment of trophic state

The assessment of the structure and functioning of an aquatic ecosystem, expressed as the deviation of the existing status from the undisturbed (reference) conditions, is the requirement set for monitoring systems by European Directive 2000/60/EC, called the Water Framework Directive [WFD; EC 2000]. Macrophytes are one of the basic elements used in addition to phytoplankton, macrozoobenthos and fish in the assessment of ecological status and classification.

The monitoring of all the biological elements, also including macrophytes, requires the development of complementary methodologies:

(i) The methodology of a field survey, i.e. the manner of material sampling;

(ii) The methodology for the water assessment based on the material collected in the field, i.e. the manner of data analysis and calculation of biological indicators.

3.1 The methodology of field survey

Transect method

The most universally applied method for aquatic vegetation surveys in both lakes and rivers is the transect-based method. In general it can be divided to simple transects and belt transects. The latter method is recommended by the European Committee for Standardisation CEN (Comité Europeén de Normalisation) [CEN 2002, 2003]. The method consists in establishing transects (sectors) perpendicular to the lake shoreline, with a length covering the complete depth range of macrophyte occurrence, and in estimating the quantitative share and sometimes also maximum colonisation depth of each species identified within a transect.

It is relatively easy and not very time-consuming to investigate the vegetation in transects. At the same time, it provides reliable and detailed information of the depth distribution of different species. For this reason, this method has been commonly applied in monitoring methodologies in many European countries.

Sampling procedure

The WISER approach for lake macrophyte survey is a modification of the transect method, which is recognized as a widely used in several European countries and compliant with CEN standard (although CEN gives only a very rough description leaving much space for interpretation and details).

Equipment used for field survey

- Plastic bags for the collection of samples, small hard plastic containers for fragile species (*Chara* sp. etc.) with additional waterproof labels
- Field protocol in sufficient copies with field pad (Appendix 1)
- Floras, relevant field guides and preliminary check list of macrophyte species
- Boat suitable for local conditions

- GPS (Global Positioning System)
- Rake with extendable rod and double-sided Luther-rake (Figure 3) with soft rope marked by depth readings

Vegetation survey within a transect

1. A transect is a belt perpendicular to the shoreline, of the length from the shoreline to the maximum depth of plant growth and of the width of approximately twice the width of the boat.

2. The position of the starting point is also the position of the first of three transects per site. The other two transects are located to the left and right, respectively, of the first transect. The minimum distance between two transects was 5 m.

3. Photos are taken in order to give a general overview of the site.

4. The starting point of the transect was situated at the beginning of the supralittoral vegetation (wetland) or at the shoreline when easily recognizable.

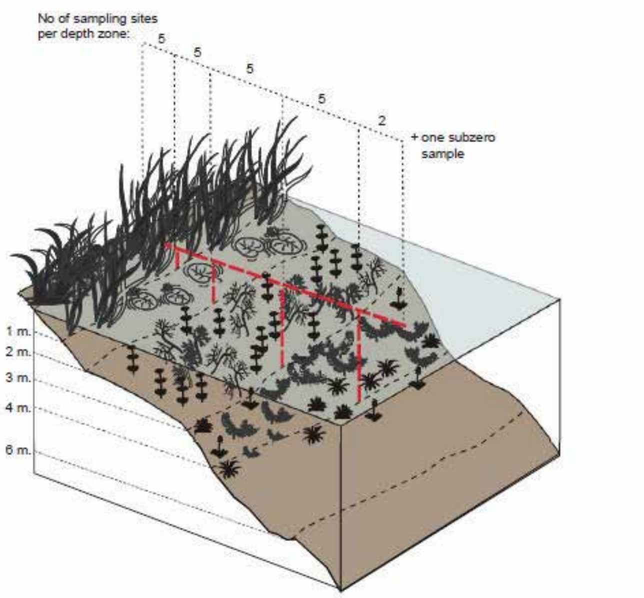

Figure 2: The scheme of phytolittoral division into depth zones and sampling site location according to the WISER belt-transect method

5. Each transect was divided into 1m depth zones with balloons as signs (Figure 5). At each depth zone, five sampling sites, evenly distributed within the zone length, were determined (according to the pattern as presented in Figure 2).

6. To assure that the maximum depth of plant growth was defined properly, at the end of transect the subzero zone was examined, i.e. a subzero sample is taken.

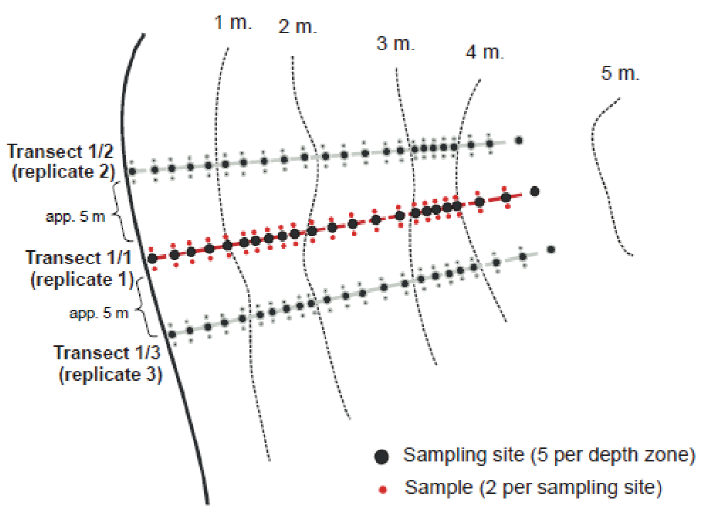

Figure 3: Sampling pattern within a sampling location (transect in three replicates)

7. Going along the transect, at each sampling site within a depth zone two macrophyte samples were taken, i.e. one sample from each side of the boat (Fig. 3).

8. The water-depth for each sample was recorded.

9. Based on two samples within a sampling site all species were identified and their abundance was estimated in according to the 5-degree scale of Melzer (1999).

10. Overall abundance of the macrophyte cover within a sampling site was also determined.

Species identification and common taxa list

• During the survey, all vascular plants including floating, leaved and submerged species as well as characeans where identified.

• Plant identification was done at species level using appropriate floras (Paparisto *et al*., 1988; Qosja et al., 1992, 1996; Vangjeli et al., 2000; Bazzichelli & Abdelahad, 2009; Casper & Krausch, 1980; Krause, 1997; Wood & Imahori, 1965, etc.).

• Specimens that could not be readily identified in the field were stored in plastic bags for further investigation. Bigger plants were dried with newspaper, smaller thin-leaved or fragile plants were preserved in alcohol.

Figure 4: Double-sided Luther-rake used for macrophyte sampling

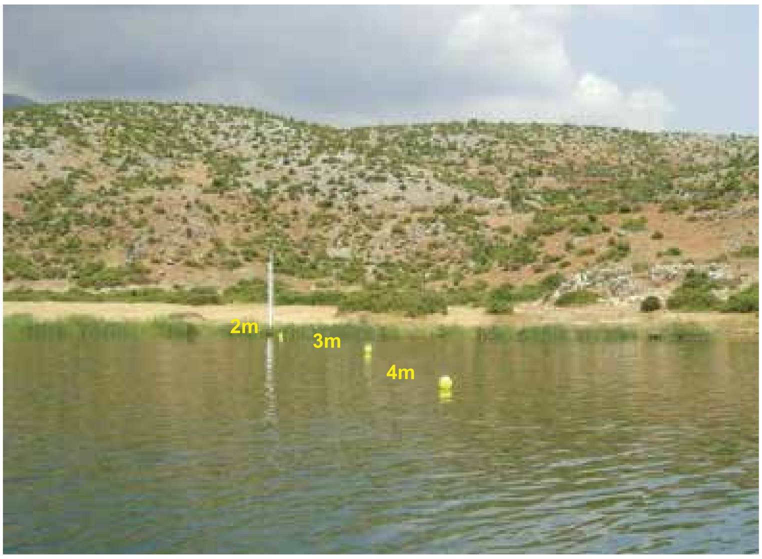

Figure 5: Placement of balloons as depth signs

Measurement of macrophyte abundance

Apart from the taxonomic composition, abundance is an important aspect of the aquatic vegetation which must be considered (under the Water Framework Directive) in macrophyte surveys. The abundance measurement scales applied in survey practice vary significantly.

In the present study, plant abundance was estimated according to a 5-degree scale: 1=very rare, 2=infrequent, 3=common, 4=frequent and 5=abundant, predominant (Melzer, 1999).

3.2 Data analysis and calculation of Macrophyte Index

All data collected at each sampling point, qualitative and quantitative, were used to calculate the Macrophyte Index according to the formula described by Melzer (1999):

$$MI = \frac{\sum_{i=1}^{n} I_i \cdot Q_i}{\sum_{i=1}^{n} Q_i}$$

where:

MI = Macrophyte Index

Ii = Indicator value of i-th species

Qi = Plant quantity of i-th species

n = Total number of species with an indicator value

To calculate the Macrophyte Index each abundance value should be cubed ($y = x^3$) because the correlation between plant abundance and quantity is not linear (Melzer, 1988; Kohler and Janauer, 1997) (Table 3).

Although the sampling follows the WISER procedure (regularly at each meter depth), MI was calculated separately for 4 different depth zones (0–1 m, 1–2 m, 2–4 m and 4 m down to the vegetation limit) and then averaged for all depths of each sampling station or belt transect according to Melzer's method. Species abundance was calculated for each depth zone as the average of the species present in the site.

Table 2: Example: calculation of MI at Sterbeq (zone depth 0-1m). The abundance values are cubed

Depth zone	Species list: Sampling sites within the depth zone	Transect 1/1					Transect 1/2					Transect 1/3					I	Q	$\sum I_i x Q_i$
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**			
0-1 m	*Ceratophyllum demersum*			1	1	1	27	8	1	1	1	1	1	1	1	1	5	46	230
	Myriophyllum spicatum				1				1	1	1				1	8	3	13	39
	Potamogeton lucens				1	8	1	1	8	8	8				1	1	3.5	37	129.5
	Potamogeton perfoliatus	1	1	1	8	27	8	8	27	8	27	1	1	1	8	27	3	154	462
	Potamogeton praelongus					8					8				1	8	3.5	25	87.5
		$\sum Q_i$																275	948

The division into depth zones must be fully respected for correct index calculation (Schaumburg et al., 2007).

Table 3: Relationship between the five-degree-scale and the quantity of submerged macrophytes

Five-degree-scale	meaning	plant quantity Q
1	very rare	1
2	infrequent	8
3	common	27
4	frequent	64
5	abundant	125

A catalogue of nine indicator groups of macrophyte species (Melzer and Schneider, 2001), exhibiting different sensitivity towards nutrient enrichment, was used to calculate the macrophyte index, ranging from 1 to 5. Species assigned to indicator group 1 are restricted to oligotrophic conditions, whereas those in indicator group 5 mainly occur in eutrophic, nutrient-rich lakes or sections of lakes. The remaining seven groups range from 1.5 to 4.5 and represent transitions between these two extremes. The assignment of macrophyte species to indicator groups was based on expert opinion and literature data. The mean MI of a lake correlates with its total phosphorus concentration during circulation time.

Table 4: The macrophyte indicator groups (Melzer and Schneider, 2001)

Indicator group 1.0	Indicator group 1.5	Indicator group 2.0
Chara hispida *Chara polyacantha* *Chara strigosa* *Potamogeton coloratus* *Utricularia stygia*	*Chara aspera* *Chara intermedia* *Utricularia minor*	*Chara delicatula* *Chara tomentosa* *Potamogeton alpinus*
Indicator group 2.5	**Indicator group 3.0**	**Indicator group 3.5**
Chara contraria *Chara fragilis* *Nitella opaca* *Nitellopsis obtusa* *Potamogeton gramineus* *Potamogeton natans* *Potamogeton x zixii*	*Chara vulgaris* *Myriophyllum spicatum* *Potamogeton filiformis* *Potamogeton perfoliatus* *Utricularia australis*	*Myriophyllum verticilatum* *Potamogeton berchtoldii* *Potamogeton lucens* *Potamogeton praelongus* *Potamogeton pusillus*
Indicator group 4.0	**Indicator group 4.5**	**Indicator group 5.0**
Hippuris vulgaris *Lagarosiphon major* *Potamogeton pectinatus*	*Elodea canadensis* *Elodea nuttallii* *Potamogeton compressus* *Potamogeton crispus* *Potamogeton obtusifolius* *Ranunculus circinatus* *Ranunculus trichophyllus*	*Ceratophyllum demersum* *Lemna minor* *Potamogeton mucronatus* *Potamogeton nodosus* *Sagittaria sagittifolia* *Spirodela polyrhiza* *Zannichellia palustris*

Six classes of the macrophyte index were designated, each representing different degrees of nutrient pollution. These six classes are assigned different colours (Table 5) to allow a clear illustration of the results.

Table 5: Relationship between index class, degree of nutrient status and assigned colour

Index class	Degree of nutrient enrichment	
1.00–1.99	Slight	Dark blue
2.00–2.49	Low	Pale blue
2.50–2.99	Moderate	Green
3.00–3.49	Immense	Yellow
3.50–3.99	Heavy	Orange
4.00–5.00	Massive	Red

4. Analysis and evaluation

4.1. Prespa Lake: Composition of macrophyte vegetation

Table 6: List of macrophyte species sampled at Prespa Lake along two transects (Gollomboc and Pustec) in summer 2013. Numbers in bold signify indicator species according to Melzer and Schneider (2001).

Nr.	Scientific name	Common name	Family	Sampling points	
				AL 1 Gollomboc	AL 2 Pustec
1	*Chara contraria*	Opposite stonewort	Characeae	x	
2	*Chara denudata*		Characeae	x	x
3	*Chara globularis*	Fragile stonewort	Characeae	x	
4	*Chara ohridana*		Characeae	x	x
5	*Nitella syncarpa*		Characeae	x	
6	*Nitellopsis obtusa*	Starry stonewort	Characeae	x	x
7	*Alisma lanceolata*		Alismataceae		x
8	*Butomus umbellatus*	Flowering rush	Butomaceae		x
9	*Ceratophyllum demersum*	Rigid hornwort	Ceratophyllaceae	x	x
10	*Eleocharis palustris*	Needle spikerush	Cyperaceae		x
11	*Elodea canadensis*	Canadian Waterweed	Hydrocharitaceae	x	x
12	*Myriophyllum spicatum*	Eurasian watermilfoil	Haloragaceae	x	x
13	*Najas marina*	Spiny Naiad	Najadaceae	x	
14	*Najas minor*	Brittle Naiad	Najadaceae	x	x
15	*Nymphoides peltata*	Water fringe	Menyanthaceae		x
16	*Persicaria amphibia*	Water knotweed	Polygonaceae	x	x
17	*Phragmites australis*	Common reed	Poaceae	x	x
18	*Potamogeton berchtoldii*	Berchtold's pondweed	Potamogetonaceae		x
19	*Potamogeton crispus*	Curly-leaf Pondweed	Potamogetonaceae	x	x
20	*Potamogeton filiformis*	Slender pondweed	Potamogetonaceae	x	x
21	*Potamogeton lucens*	Shining pondweed	Potamogetonaceae	x	x
22	*Potamogeton nodosus*	Longleaf pondweed	Potamogetonaceae		x
23	*Potamogeton perfoliatus*	Perfoliate pondweed	Potamogetonaceae	x	x
24	*Potamogeton pusillus*	Small pondweed	Potamogetonaceae	x	x
25	*Schoenoplectus lacustris*	Common Club-rush	Cyperaceae		x
26	*Trapa natans*	Water chestnut	Trapaceae		x
27	*Vallisneria spiralis*	Straight Vallisneria	Hydrocharitaceae	x	x
28	*Zannichellia palustris*	Horned pondweed	Potamogetonaceae	x	

A total number of 28 aquatic macrophyte species were recorded at Lake Prespa, 20 species at Gollomboc and 23 species at Pustec. Twelve species at Gollomboc and 11 species at Pustec have an indicator values ranging from 2.5 (minimum) to 5.0 (maximum) (Tables 6 and 7).

The most commonly encountered species were *Ceratophyllum demersum* and *Myriophyllum spicatum*, which are both listed as macrophyte indicators.

The maximum depth of plant growth recorded at both station was 5.8m.

MI (Macrophyte index) calculation

The Macrophytes Index (MI) calculated varied from 3.85 (Gollomboc) to 3.90 (Pustec) which correspond with "heavy" nutrient enrichment.

Gollomboc – MI average all depths = 3.85 (Orange)

MI according to the 4 different depth zones

Depth zone 0-1m MI= 3.78

Depth zone 1-2m MI= 3.15

Depth zone 2-4m MI= 4.30

Depth zone 4-6m MI= 4.18

Pustec – MI average all depths = 3.90 (Orange)

MI according to the 4 different depth zones

Depth zone 0-1m MI= 3.24

Depth zone 1-2m MI= 3.20

Depth zone 2-4m MI= 4.55

Depth zone 4-6m MI= 4.64

At both sampling points, a considerable number of species was found, 20 and 23 species respectively, and the MIs show almost the same trophic state

The two sampling points are located in residential areas, but apparently, Pustec is more impacted by human activity. Pustec village is more populated than Gollomboc and the land is more cultivated and closer to the sampling point.

4.2. Shkodra Lake

Composition of macrophyte vegetation

The total number of aquatic macrophyte species recorded at Shkodra Lake was 15, notably: Sterbeq 14 species, Zogaj 9 species and Shiroka 10 species. Eight of these species are macrophyte indicators according to Melzer and Schneider (2001), the indicator values ranging from 2.5 (minimum) to 5.0 (maximum). The maximum depth of plant growth varied from 5.3m in Shiroka to 6.7m in Sterbeq. Dominant species were *Ceratophyllum demersum, Potamogeton perfoliatus* and *Potamogeton lucens.*

Table 7: List of macrophyte species sampled at Shkodra/Skadar Lake along three transects (Sterbeq, Zogaj and Shiroka) in summer 2013. Numbers in bold signify indicator species according to Melzer and Schneider (2001).

No.	Scientific name	Common name	Family	Sample points		
				AL 1 Sterbeq	AL 2 Zogaj	AL 3 Shiroka
1	*Nitellopsis obtusa*	Starry stonewort	Characeae	x		
2	*Ceratophyllum demersum*	Rigid hornwort	Ceratophyllaceae	x	x	x
3	*Gratiola officinalis*	Common hedgehyssop	Plantaginaceae	x		
4	*Juncus sp.*	Rush	Juncaceae	x		
5	*Myriophyllum spicatum*	Eurasian watermilfoil	Haloragaceae	x	x	x
6	*Najas marina*	Spiny Naiad	Najadaceae	x	x	x
7	*Najas minor*	Brittle Naiad	Najadaceae	x	x	x
8	*Paspalum distichum*	Knotgrass	Poaceae	x		
9	*Potamogeton crispus*	Curly-leaf Pondweed	Potamogetonaceae	x	x	x
10	*Potamogeton lucens*	Shining pondweed	Potamogetonaceae	x		x
11	*Potamogeton nodosus*	Longleaf pondweed	Potamogetonaceae		x	x
12	*Potamogeton perfoliatus*	Perfoliate pondweed	Potamogetonaceae	x	x	x
13	*Potamogeton praelongus*	White-stem Pondweed	Potamogetonaceae	x	x	x
14	*Potamogeton pusillus*	Slender pondweed	Potamogetonaceae	x		
15	*Vallisneria spiralis*	Straight Vallisneria	Hydrocharitaceae	x	x	x

MI (Macrophyte index) calculation

The calculated Macrophyte Indices (MI) indicated heavy nutrient enrichment for Sterbeq (3.52) and Shiroka (3.80) and massive nutrient enrichment for Zogaj (4.3).

Sterbeq – MI average all depths = 3.68 (Orange)

MI according to the 4 different depth zones

Depth zone 0-1m	MI= 3.44
Depth zone 1-2m	MI= 3.14
Depth zone 2-4m	MI= 3.53
Depth zone 4-7m	MI= 4.61

Zogaj – MI average all depths = 4.3 (Red)

MI according to the 4 different depth zones

Depth zone 0-1m	MI= 4.00
Depth zone 1-2m	MI= 3.37
Depth zone 2-4m	MI= 4.92
Depth zone 4-6m	MI= 5.00

Shiroke – MI average all depths = 3.86 (Orange)

MI according to the 4 different depth zones

Depth zone 0-1m	MI= 3.38
Depth zone 1-2m	MI= 4.00
Depth zone 2-4m	MI= 3.58
Depth zone 4-6m	MI= 4.50

According to previous studies, the values of chlorophyll concentration and TSI indicate that the waters of the lake are mesotrophic with a tendency towards eutrophic conditions during summer. The present results lead more or less to the same conclusion.

In the northeastern part of the lake (sampling point Sterbeq) the shore is flat, gradually descending towards the lake's bottom and often with rich underwater vegetation. Rising water levels of the lake in winter and spring may cause the accumulation of nutrients from cultivated land.

Nutrient enrichment and the subsequent increase of aquatic vegetation result from solid organic matter washed into the lake during flooding, soil from the deforested steep slopes around the lake and from large agricultural fields on the north and east side.

The south-west coast (sampling points Zogaj, Shiroka) shows a different picture: It is steep and rocky, no streams flow into the lake and there is little cultivated land. It is likely that the higher levels of trophic state can be explained mainly by human impact.

All three sampling stations are located near to human settlements (villages) characterized by variable human activities such as intensive agriculture, farming, fishing, tourism accompanied also by rapid urbanization. There are about 1,000 residents and a series of bars and restaurants (about 22) from Zogaj to Shiroka (Bejko, 2012).

The human activities, including discharges of sewage and other organic materials with phosphorus (detergents); inappropriate use of chemical fertilizers, which drain into the lake basin karsts formations; massive cutting of forests and lack of anti-erosion measures in the watershed of Lake Shkodra, which has to do with the input of solid organic matter, impact the quality of water.

Table 8: Summary of data and degree of nutrient enrichment for all sampling points at Lakes Prespa and Shkodra (Albanian side, summer, 2013)

Lake	Sampling point	No. of species recorded	No. of species with an indicator value	Max. depth of plants growth	MI	Degree of nutrient enrichment
Prespa	AL 1 Gollomboc	20	12	5.8	3.85	Heavy
	AL 2 Pustec	23	11	5.8	3.90	Heavy
Shkodra	AL 1 Sterbeq	14	7	6.7	3.68	Heavy
	AL 2 Zogaj	9	6	5.5	4.30	Massive
	AL 3 Shiroka	10	8	5.3	3.86	Heavy

Figure 6: Maps showing sampling stations and degree of nutrient enrichment at Shkodra Lake and Prespa Lake (Albanian side, summer 2013)

5. Summary and recommendations

Aquatic plants have an important role in biological monitoring of natural habitats because changes in aquatic species composition and abundance are closely linked with water, sediment, and surrounding land quality.

In August 2013 a macrophyte survey was conducted at Lakes Shkodra and Prespa in the frame of the "Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar (CSBL)" Project.

The overall aim of the survey was to estimate the ecological status of the lakes using macrophytes as biological quality elements corresponding to the requirements of the European Water Framework Directive (WFD).

Three transects were sampled at Shkodra Lake and two transects at Prespa Lake. At each point, a survey of macrophyte species was performed along a belt transect according to the WISER method. All data collected, qualitative and quantitative in nature, were used to calculate the Macrophyte Index (Melzer, 1999).

Twenty-eight aquatic macrophyte species were recorded at Prespa Lake (Gollomboc 20 species, Pustec 23 species) and 15 at Shkodra Lake (Sterbeq 14 species, Zogaj 9 species and Shiroka 10 species).

Due to the high degree of natural variability among the emerged vegetation, only submerged macrophytes (hydrophytes) were used to assess the nutrient status of the lakes.

The results indicated that both lakes are affected by human activities. The nutrient enrichment of Prespa Lake was heavy at both sites (Gollomboc and Pustec). At Lake Shkodra, nutrient enrichment was heavy at two sites (Sterbeq and Shiroka) and massive at one site (Zogaj).

However, these data cannot indicate the ecological state of the whole lake, because the number of transects should be much higher. Considering the recommended number of transects in relation to lake surface (Schaumburg et al., 2007), the number of transects should be 30-50 for lakes with a surface area of more than 100km^2.

Since Shkodra Lake lies in the sub-Mediterranean climate zone, the macrophyte index method (Melzer's method, 1999) used for this lake needs to be adapted to Mediterranean limnosystems. As a consequence of the regionally differing behaviour of some species, an overall application of the Macrophyte Index is presumably not possible without screening of the composition of characteristic macrophyte communities and reevaluating and/or extending the existing indicator group classification scheme (Melzer, 1999).

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7. Appendix 1. Field protocols

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE					Page 2/4
MACROPHYTE SURVEY					
Transect no: 1	**Gollomboc**	**Transect 1/1**	**Transect 1/2**	**Transect 1/3**	
Vegetation spatial structure within a transect					
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids					
Growth form dominating		ce	ce	ce	
Start- point depth (m)		0.2	0.2	0.2	
End-point depth (m)		7.5	7.5	7.5	
Maximum depth of plant growth (m)		5.8 m	5.6 m	5.6 m	
Max. observed depth of rush (m)		-	-	-	
Max. observed depth of elodeids (m)		5.2	5.8	5.2	
Max. observed depth of charids (m)		5.8	5.0	5.5	
Max. observed depth of isoetids (m)		-	-	-	
Bottom quality: *total cover of whole zone as percentage*					
Rock (>4000 mm)					
Boulders (250-4000 mm)					
Stone (16-250 mm)					
Gravel (2-16 mm)		x	x	x	
Sand (0.06-2 mm,)		x	x	x	
Silt (smooth between fingers)					
Clay (elastic, grey)					
Mud		x	x	x	
Peat					
Detritus (tree leaves, trash, etc.)					
Other (specify)					

Species composition on a belt transect (abundance)																
Depth zone	Species list: Sampling sites within the depth zone	**Transect 1/1**					**Transect 1/2**					**Transect 1/3**				
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
0 - 1 m continued	Chara globularis				1	2								1	1	
	Chara ohridana	1	2									1	1			
	Ceratophyllum demersum				1	1								1		
	Elodea canadensis															1
	Myriophyllum spicatum	1			1	1				1	1	1	1	1	2	3
	Najas minor	1		1												
	Persicaria amphibia	1										1		1	2	3
	Potamogeton crispus	1														
	Potamogeton filiformis	1		1								1	1	1		
	Potamogeton lucens					1										
	Potamogeton perfoliatus									1	1	1	1	1		2
	Vallisneria spiralis	1	1									1	1			
	Zannichellia palustris	2	2	1		1						2	2	1		
1 - 2 m	Chara ohridana	1														
	Ceratophyllum demersum				1	1			1	1	1	1			1	1
	Elodea canadensis			1	1	1						1			1	2
	Myriophyllum spicatum	1	1	4	2	1		4	3	3	2	3	3	3	2	3
	Najas marina														1	1
	Persicaria amphibia											1	5	3		
	Phragmites australis		1										2	2		
	Potamogeton filiformis	1	1		1	1										
	Potamogeton lucens							2	1							
	Potamogeton perfoliatus	1						2	1		1					
	Potamogeton pusillus							1								
	Zannichellia palustris							2								
2 - 3m	Chara contraria								1							
	Chara globularis										1					
	Nitellopsis obtusa							1	1							
	Ceratophyllum demersum	1	1	1	1	3	1	2	3	3	3		2	2	2	2
	Elodea canadensis	1	1	1				1	1	1		1	1			
	Myriophyllum spicatum	2	2	1		1	1	1	1	1	2	2	2	2	1	2
	Potamogeton filiformis			1	1				1			1			1	
	Potamogeton perfoliatus		1	1	3	1	1	1	1				2	2	3	
	Potamogeton pusillus					1					1	1		1		1
3 - 4 m	Chara denudata														1	1
	Nitellopsis obtusa		1	2		2	2	1	1	1	2	1	2			
	Ceratophyllum demersum	2	2	2	1	3	3	3	3	3	4	3	3	2	2	2
	Elodea canadensis		1	1										1	1	1
	Myriophyllum spicatum	1	1		1	1	1	2	1	1	1	1		1	1	1
	Potamogeton filiformis			1	1											
	Potamogeton pusillus		1			1		1		1	1	1		1		
4 - 5m	Chara denudata	1											1			
	Nitellopsis obtusa	2	1	2	1	2	1	1	1	1	2	1	2	1	1	
	Nitella syncarpa											1	1			
	Ceratophyllum demersum	2	1	1	1	1	3	2	1	1	1	2	3	3	2	1
	Myriophyllum spicatum						1	1			1					
	Potamogeton pusillus	1		1	1					1			1			
5 - 6m	Nitellopsis obtusa	1	1	1	1							1	1	1		
	Ceratophyllum demersum	1	1	1	1		1	1	1	1		1	1	1		
	Elodea canadensis							1	1	1						
	Najas marina		1													
	Potamogeton pusillus												1			
6 - 7m	**No plants**															

<table>
<tr><td colspan="4">FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE</td><td rowspan="2">Page 1/4</td></tr>
<tr><td colspan="4">GENERAL INFORMATION</td></tr>
<tr><td>Lake name:
Prespa</td><td>Lake type:</td><td>Lake code:</td><td>Surveyor: L. Kashta, M. Rakaj</td><td>Date:
23.7.2013</td></tr>
<tr><td colspan="5">Lake information</td></tr>
<tr><td>Surface area (km²)</td><td></td><td colspan="2">Lake altitude (m asl)</td><td></td></tr>
<tr><td>Maximum depth (m)</td><td></td><td colspan="2">Mean alkalinity (meq)</td><td></td></tr>
<tr><td>Mean depth (m)</td><td></td><td colspan="2">Water colour (mg/Pt/L)</td><td></td></tr>
<tr><td>Lake perimeter incl. islands (km)</td><td></td><td colspan="2">Dominating geology in catchment
(S=siliceous, C=calcareous, O=organic)</td><td></td></tr>
<tr><td>Retention time</td><td></td><td colspan="2">Dominating landuse in cathcment
(F=forest, A=agricultural, U=urban, O=other)</td><td></td></tr>
<tr><td colspan="5">P of locality</td></tr>
<tr><td>Locality number:</td><td colspan="2">Starting time:</td><td colspan="2">Ending time:</td></tr>
<tr><td>Start-point (GPS)</td><td colspan="2"></td><td colspan="2"></td></tr>
<tr><td>End-point (GPS)
40⁰ 47’ 27.2” N
20⁰ 54’ 42.2“ E</td><td colspan="2"></td><td colspan="2"></td></tr>
<tr><td>Locality direction (degrees)</td><td colspan="4"></td></tr>
<tr><td colspan="5">Freehand drawing of transect</td></tr>
<tr><td colspan="5"></td></tr>
</table>

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 2/4
MACROPHYTE SURVEY				
Transect no: 2	**Pustec**	**Transect 2/1**	**Transect 2/2**	**Transect 2/3**
Vegetation spatial structure within a transect				
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids				
Growth form dominating		ce	ce	ce
Start- point depth (m)		0.1	0.1	0.1
End-point depth (m)		7.5	7.5	7.5
Maximum depth of plant growth (m)		5.8	5.7	5.7
Max. observed depth of rush (m)		1.8	1.8	2.0
Max. observed depth of elodeids (m)		5.4	5.5	5.7
Max. observed depth of charids (m)		5.5	5.4	5.5
Max. observed depth of isoetids (m)		-	-	-
Bottom quality: *total cover of whole zone as percentage*				
Rock (>4000 mm)				
Boulders (250-4000 mm)				
Stone (16-250 mm)				
Gravel (2-16 mm)		x	x	x
Sand (0.06-2 mm,)				
Silt (smooth between fingers)				
Clay (elastic, grey)				
Mud		x	x	x
Peat				
Detritus (tree leaves, trash, etc.)				
Other (specify)				

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE					Page 2/4
MACROPHYTE SURVEY					
Transect no: 2	**Pustec**	**Transect 2/1**	**Transect 2/2**	**Transect 2/3**	
Vegetation spatial structure within a transect					
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids					
Growth form dominating		ce	ce	ce	
Start- point depth (m)		0.1	0.1	0.1	
End-point depth (m)		7.5	7.5	7.5	
Maximum depth of plant growth (m)		5.8	5.7	5.7	
Max. observed depth of rush (m)		1.8	1.8	2.0	
Max. observed depth of elodeids (m)		5.4	5.5	5.7	
Max. observed depth of charids (m)		5.5	5.4	5.5	
Max. observed depth of isoetids (m)		-	-	-	
Bottom quality: *total cover of whole zone as percentage*					
Rock (>4000 mm)					
Boulders (250-4000 mm)					
Stone (16-250 mm)					
Gravel (2-16 mm)		x	x	x	
Sand (0.06-2 mm,)					
Silt (smooth between fingers)					
Clay (elastic, grey)					
Mud		x	x	x	
Peat					
Detritus (tree leaves, trash, etc.)					
Other (specify)					

Species composition on a belt transect (abundance)																
Depth zone	Species list: Sampling sites within the depth zone	**Transect 2/1**					**Transect 2/2**					**Transect 2/3**				
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
0 - 1 m continued	Chara ohridana					1					2					
	Alisma lanceolata	1										2				
	Ceratophyllum demersum	1	1	1	1	1		1	1	1	1					
	Butomus umbellatus	2	4		1		1	1	1	2	1		1		2	
	Eleocharis palustris														3	2
	Elodea canadensis	1	1	1	1	1		1	1	1	1					
	Myriophyllum spicatum	4		2	2	4	2	2	1	2	2	2	2		1	2
	Najas minor	1	1													
	Nymphoides peltata		1									1				
	Persicaria amphibia	1	1						1	1			1			
	Phragmites australis					2							1			
	Potamogeton berchtoldii	1														
	Potamogeton crispus	1														
	Potamogeton filiformis	1						1	1	1	1		1			
	Potamogeton lucens		1	1	1	1										
	Potamogeton nodosus		1					1	1	1			2			
	Potamogeton perfoliatus	1				1										
	Potamogeton pusillus		1				1									
	Schoenoplectus lacustris		1	1								1				
	Trapa natans	1	1				1									
	Vallisneria spiralis	4	2	2	1		2	1	1	1	1	3	1		1	1
1 - 2 m	Chara ohridana	1					2	1	1		1					
	Chara denudata															1
	Butomus umbellatus	2														
	Ceratophyllum demersum	2	2	1		2	1	1				1	1		1	1
	Elodea canadensis				1	1		1	1		1	1	1	1		
	Myriophyllum spicatum	3	3	4	3	3	2	3	3	3	3	3	2	2	1	2
	Phragmites australis				2							2	2			
	Potamogeton filiformis											1				
	Potamogeton lucens		1	2								1	1			
	Potamogeton perfoliatus		1	1												
	Schoenoplectus lacustris			2	3		2	3	3	3		1	1	2	3	3
	Trapa natans		1			1			1	1		1		1		1
2 - 3 m	Nitellopsis obtusa											1	2	2	1	1
	Ceratophyllum demersum	2	2	4	3	4	2	2	2	2	2	3	3	2	2	2
	Elodea canadensis	1	1	1	3	2		2	1		4	2	1	2	1	3
	Myriophyllum spicatum	3	2	2	1	2	4	3	2	2	2	2	2	1	1	
	Potamogeton pusillus	1		1	1	1	1		1	1			1		1	2
	Trapa natans	1	1			1						1	1			
	Vallisneria spiralis											1	1			
3 - 4 m	Nitellopsis obtusa	1	1										1			1
	Ceratophyllum demersum	3	3	4	5	4	2	2	3	5	4	4	3	4	5	4
	Elodea canadensis	3	3	3	3	1	1	3	2	1	2	3	3	2	1	1
	Myriophyllum spicatum	1	3	1	1	1	1	1	1	1	1	1	1	1	2	
	Potamogeton pusillus	1												1	1	
	Trapa natans	1														
4 - 5m	Nitellopsis obtusa		1	1	1	1										
	Ceratophyllum demersum	3	3	3	3	2	3	3	4	3	2	4	4	3	3	2
	Elodea canadensis	1	1	1			1	1				1	1	1		
	Myriophyllum spicatum	1	1	1			1	1	1			1	1	1	1	
	Potamogeton pusillus				1	2	1	1					1			1
5 - 6m	Nitellopsis obtusa	2	2	1			2	1				1	2	2		
	Ceratophyllum demersum	2	1	1	1	1	1	3	2	1		1	2	1	1	
	Myriophyllum spicatum							1	1			1	1		1	
	Potamogeton pusillus	1	1													
6 - 7m	**No plants**															

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 1/4
GENERAL INFORMATION				
Lake name: Shkodra/Scadar	**Lake type:**	**Lake code:**	**Surveyor:** L. Kashta, M. Rakaj	**Date:** 11.8.2013

Lake information			
Surface area (km^2)		Lake altitude (m asl)	
Maximum depth (m)		Mean alkalinity (meq)	
Mean depth (m)		Water colour (mg/Pt/L)	
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)	
Retention time		Dominating landuse in cathcment (F=forest, A=agricultural, U=urban, O=other)	

P of locality		
Locality number: AL 1	**Starting time:**	**Ending time:**
Start-point (GPS) 42°11'45.8" N 19°23'21.8" E		
End-point (GPS)		
Locality direction (degrees)		

Freehand drawing of transect

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE					Page 2/4
MACROPHYTE SURVEY					
Transect no: 1	**Sterbeq**	**Transect 1/1**	**Transect 1/2**	**Transect 1/3**	
Vegetation spatial structure within a transect					
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids					
Growth form dominating		el	el	el	
Start- point depth (m)		0.2	0.2	0.2	
End-point depth (m)		7	7	7	
Maximum depth of plant growth (m)		6.6	6.7	6.6	
Max. observed depth of rush (m)		-	-	-	
Max. observed depth of elodeids (m)		6.6	6.5	6.6	
Max. observed depth of charids (m)		3	3	3.3	
Max. observed depth of isoetids (m)		-	-	-	
Bottom quality: *total cover of whole zone as percentage*					
Rock (>4000 mm)					
Boulders (250-4000 mm)					
Stone (16-250 mm)					
Gravel (2-16 mm)					
Sand (0.06-2 mm,)					
Silt (smooth between fingers)					
Clay (elastic, grey)					
Mud					
Peat					
Detritus (tree leaves, trash, etc.)					
Other (specify)					

Species composition on a belt transect (abundance)																
Depth zone	Species list: Sampling sites within the depth zone	**Transect 1/1**					**Transect 1/2**					**Transect 1/3**				
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
0 - 1 m continued	Ceratophyllum demersum			1	1	1	3	2	1	1	1	1	1	1	1	1
	Gratiola officinalis	2	2				1	1				3	3	3		
	Juncus sp.	1	1	1			1					1				
	Myriophyllum spicatum				1				1	1	1				1	2
	Najas marina	1	1		4	1				3	2				3	1
	Najas minor	1	1	2	3	2	1	1	1	2	1	1	1	2	2	2
	Paspalum distichum	2										1				
	Potamogeton lucens				1	2	1	1	2	2	2				1	1
	Potamogeton perfoliatus	1	1	1	2	3	2	2	3	2	3	1	1	1	2	3
	Potamogeton praelongus					2					2				1	2
	Vallisneria spiralis	1	1	1		1	3	2	2		1	1	2	3	1	1
1 - 2 m	Ceratophyllum demersum	1	1	1			1	2				1	1	2	1	1
	Myriophyllum spicatum			1		1		1	1		1			1		2
	Najas marina		1	2	1	2		2	2	2	3		2	2	1	2
	Najas minor		1	1	1	1	1	3	2	1	2		1	1	1	1
	Potamogeton lucens		1	2	2	1		2	2	2	1			2	2	2
	Potamogeton perfoliatus	1	2	4	3	2		3	2	3	2	1	2	3	3	2
	Vallisneria spiralis	1		1	1	1	1	1	1	1	1	1	1	1	1	1
2 - 3m	Nitellopsis obtusa					1				1	1					
	Ceratophyllum demersum	1	1	1	1	1		2	1	1	1	1	2	1	1	1
	Myriophyllum spicatum		1	1	1	1		1	1	1	1			1	1	1
	Najas marina	1	2	1	2	1		2	1	1	2	2	2	2	1	1
	Najas minor	1	1		3	4		2	2	3	4	1	1	4	3	3
	Potamogeton lucens		2	1	2	2		2	2	2	2	2	2	1	2	3
	Potamogeton perfoliatus		1		1	2		2	1	1	2		2	1	2	2
	Potamogeton pusillus					1				1	1					1
	Vallisneria spiralis	1	1	1	2	2		2	2	2	2	2	2	2	1	1
3 - 4 m	Nitellopsis obtusa											1	1			
	Ceratophyllum demersum			1	1	1			1	1	1		1	2		1
	Myriophyllum spicatum	1	1	1	1		2	2		1		1	1	1		
	Najas marina	1	1	1	2	3	2	2	2	2	4	2	2	2		1
	Najas minor	2	3	2	3	2	3	2	2	3	2	3	2	2		2
	Potamogeton lucens	1	2	2			2					2		1		
	Potamogeton perfoliatus		1	1	1	1	1	1		1	1	1	1	1		1
	Potamogeton pusillus		1	1			1					1				
	Vallisneria spiralis	1	2	2	2	2	2	2	1		1	2	2	2		2
4 - 5m	Ceratophyllum demersum	1	2	1	1	2	1	1	2	2	2	2	2	1	1	2
	Najas marina	2	4	3	3	3		3	4	3	3	3	3	2	2	2
	Najas minor	1	2	2	1	1	1	1	1		2	1	2	1	1	1
	Potamogeton lucens								1	1	1		1	1	1	1
	Potamogeton perfoliatus		1		1	1			1	1	1	1	1			1
	Vallisneria spiralis	1	1	1	1	1		1	1	1	1	1	1		1	1
5 - 6m	Ceratophyllum demersum	1	2	1	1	1		3	2	2	2	2	2	3	2	2
	Najas marina	2	3	2	3	2		3	2	3	2	2	2	3	2	2
	Najas minor		1	1	1				1	1	2	1	1	1	1	
	Vallisneria spiralis				1	1		1	1	1	1	1	1		1	
6 - 7m	Ceratophyllum demersum	1	1	1	1		1	1	1					1	1	
	Najas minor		1	1	1		1	1	1			1			1	
	Najas marina											1	1	2		
	Potamogeton crispus	1														
	Vallisneria spiralis												1	1		

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				**Page 1/4**
GENERAL INFORMATION				
Lake name: **Lake Shkodra/Scadar**	**Lake type:**	**Lake code:**	**Surveyor:** L. Kashta, M. Rakaj	**Date:** 13.8.2013

Lake information			
Surface area (km²)		Lake altitude (m asl)	
Maximum depth (m)		Mean alkalinity (meq)	
Mean depth (m)		Water colour (mg/Pt/L)	
Lake perimeter incl. islands (km)		Dominating geology in catchment (S=siliceous, C=calcareous, O=organic)	
Retention time		Dominating land use in cathcment (F=forest, A=agricultural, U=urban, O=other)	

P of locality		
Locality number: AL 2	**Starting time:**	**Ending time:**
Start-point (GPS) 42° 04' 22.6" N 19° 24' 13.9" E		
End-point (GPS)		
Locality direction (degrees)		

Freehand drawing of transect

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE					Page 2/4
MACROPHYTE SURVEY					
Transect no: 2	**Zogaj**	**Transect 2/1**	**Transect 2/2**	**Transect 2/3**	
Vegetation spatial structure within a transect					
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids					
Growth form dominating		el	el	el	
Start- point depth (m)		0.2	0.2	0.2	
End-point depth (m)		6	6	6	
Maximum depth of plant growth (m)		5.5	5.5	5.5	
Max. observed depth of rush (m)		-	-	-	
Max. observed depth of elodeids (m)		5.5	5.5	5.5	
Max. observed depth of charids (m)		-	-	-	
Max. observed depth of isoetids (m)		-	-	-	
Bottom quality: *total cover of whole zone as percentage*					
Rock (>4000 mm)					
Boulders (250-4000 mm)					
Stone (16-250 mm)					
Gravel (2-16 mm)					
Sand (0.06-2 mm,)					
Silt (smooth between fingers)					
Clay (elastic, grey)					
Mud					
Peat					
Detritus (tree leaves, trash, etc.)					
Other (specify)					

Species composition on a belt transect (abundance)																
Depth zone	Species list: Sampling sites within the depth zone	**Transect 2/1**					**Transect 2/2**					**Transect 2/3**				
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
0 - 1 m continued	Myriophyllum spicatum	1		1	1				1	1		1				2
	Najas marina	1		2	2		1	1	2	2		1		2	2	2
	Najas minor		1	1	1	1		1	1	1	1			1	1	1
	Potamogeton crispus			1				1	1					1		
	Potamogeton nodosus			1		2			2					1		
	Potamogeton praelongus			1		1			1			1		1		1
	Vallisneria spiralis	1	2	2	2	3		2	3	3	3	1	2	2	3	3
1 - 2 m	Myriophyllum spicatum			2		1			1		1				2	1
	Najas marina			2		2		2		2	2	2		2	2	
	Najas minor		1		1			1	1	1			1	1	1	
	Potamogeton nodosus			1	1				1						1	
	Potamogeton praelongus	1	1			1	1		1		1	1		1		
	Vallisneria spiralis	2	3	3		3	1	2	2	3	2	1	2	2	2	
2 - 3m	Ceratophyllum demersum		1	2		1		2	3	2	3	2	2	2		1
	Najas marina		1		1				1		2			1	2	2
	Potamogeton perfoliatus			1	1				1				1			
	Vallisneria spiralis	2	2	2				2		2	1		2		2	1
3-4m	Ceratophyllum demersum	2		2		2	2	2	2		2	3	2	2		1
	Najas marina	3	2	3		3		3		2	2		3	3	4	3
	Vallisneria spiralis	1	1		1		1		1	1		1	1			
4 - 5m	Ceratophyllum demersum	1		1	1			1	1		1		1	1	1	
	Najas marina	3	2	3		3	3	3		4		3	3	4	3	4
	Vallisneria spiralis	1		1				1		1	1		1	1		
5 -5.5m	Ceratophyllum demersum		1	1			1		1			1		1		
	Najas marina	3		3			3	2	3			3	3	2		
6m	No plants															

<table>
<tr><td colspan="4">FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE</td><td rowspan="2">**Page 1/4**</td></tr>
<tr><td colspan="4">GENERAL INFORMATION</td></tr>
<tr><td>**Lake name:**
Lake Shkodra/Scadar</td><td>**Lake type:**</td><td>**Lake code:**</td><td>**Surveyor: L. Kashta, M. Rakaj**</td><td>**Date:**
12.8.2013</td></tr>
</table>

<table>
<tr><td colspan="4">***Lake information***</td></tr>
<tr><td>Surface area (km²)</td><td></td><td>Lake altitude (m asl)</td><td></td></tr>
<tr><td>Maximum depth (m)</td><td></td><td>Mean alkalinity (meq)</td><td></td></tr>
<tr><td>Mean depth (m)</td><td></td><td>Water colour (mg/Pt/L)</td><td></td></tr>
<tr><td>Lake perimeter incl. islands (km)</td><td></td><td>Dominating geology in catchment
(S=siliceous, C=calcareous, O=organic)</td><td></td></tr>
<tr><td>Retention time</td><td></td><td>Dominating land use in cathcment
(F=forest, A=agricultural, U=urban, O=other)</td><td></td></tr>
</table>

<table>
<tr><td colspan="3">***P of locality***</td></tr>
<tr><td>**Locality number: AL 3**</td><td>**Starting time:**</td><td>**Ending time:**</td></tr>
<tr><td>Start-point (GPS)
42⁰ 03' 41.3'' N
19⁰ 27' 13.9'' E</td><td></td><td></td></tr>
<tr><td>End-point (GPS)</td><td></td><td></td></tr>
<tr><td>Locality direction (degrees)</td><td colspan="2"></td></tr>
<tr><td colspan="3">***Freehand drawing of transect***</td></tr>
<tr><td colspan="3"></td></tr>
</table>

FIELD PROTOCOL FOR SURVEYING AQUATIC MACROPHYTE				Page 2/4
MACROPHYTE SURVEY				
Transect no: 3	**Shiroka**	**Transect 3/1**	**Transect 3/2**	**Transect 3/3**
Vegetation spatial structure within a transect				
GROWTH FORM TYPES: he = helophytes, ny = nymphaeids, el = elodeids, le = lemnids , ce = ceratophyllids , is = isoetids, ch = charids				
Growth form dominating		el	el	el
Start- point depth (m)		0.2	0.2	0.2
End-point depth (m)		6	6	6
Maximum depth of plant growth (m)		5.3	5.2	5.3
Max. observed depth of rush (m)		-	-	-
Max. observed depth of elodeids (m)		4.8	4.8	4.8
Max. observed depth of charids (m)		-	-	-
Max. observed depth of isoetids (m)		-	-	-
Bottom quality: *total cover of whole zone as percentage*				
Rock (>4000 mm)				
Boulders (250-4000 mm)		x	x	x
Stone (16-250 mm)		x	x	x
Gravel (2-16 mm)				
Sand (0.06-2 mm,)				
Silt (smooth between fingers)				
Clay (elastic, grey)				
Mud		x	x	x
Peat				
Detritus (tree leaves, trash, etc.)				
Other (specify)				

Species composition on a belt transect (abundance)																
Depth zone	Species list: Sampling sites within the depth zone	**Transect 3/1**					**Transect 3/2**					**Transect 3/3**				
		1	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**	**1**	**2**	**3**	**4**	**5**
0 - 1 m continued	Ceratophyllum demersum			1	1	1		1		1	1		1		1	1
	Myriophyllum spicatum				1	2				1	2				1	2
	Najas marina			1	2	1			1	2	2		1	1	2	2
	Najas minor				1	1					1			1		1
	Potamogeton crispus		1					1	1				1			
	Potamogeton lucens				1					1						
	Potamogeton nodosus		1					1	1				1			
	Potamogeton perfoliatus				1	2				2	2				2	2
	Potamogeton praelongus		1					1								
	Vallisneria spiralis	1	3	2	3		1	3	2	3		1	2	2	3	1
1 - 2 m	Ceratophyllum demersum		2	3				2	3		2	3		2	3	
	Myriophyllum spicatum			1					1	2			1		1	
	Najas marina		1		2				1	2				2	2	
	Najas minor			1	1			1	1					1	1	
	Potamogeton lucens	1	1			1	1					1	1			
	Potamogeton perfoliatus				3	3				2	3			2	3	
	Vallisneria spiralis			2	1				3	3			3	2		
2 - 3m	Ceratophyllum demersum		1	2	1			1	1	2				1	2	
	Myriophyllum spicatum			1					1					1		
	Najas marina		2	4	4			2	4	5			4	3	4	5
	Najas minor		2		2			2		2				2	2	
	Potamogeton lucens					1					1					1
	Potamogeton perfoliatus				2	2				2	3			2		3
	Vallisneria spiralis		1		2			1	2				2	1		
3 - 4 m	Ceratophyllum demersum	1	2		1		1	2		2	2	2		1	2	2
	Myriophyllum spicatum			1						1				1		
	Najas marina		2		3				2	3				3	3	2
	Najas minor			2	2					2				2	2	
	Potamogeton lucens		1	1		1	1					1		1	1	
	Potamogeton perfoliatus			2	3	3			3	4			3	3		2
	Vallisneria spiralis			2	1			1			2		1	1		
4 - 5m	Ceratophyllum demersum	1	2		1			3	1	1	1			1	2	2
	Najas marina		1		3	1			3	2				2	2	2
	Najas minor		2	3	2				1	2			2	2	1	
	Potamogeton lucens			1	1	1	1					1		1		
	Potamogeton perfoliatus		1					1	1	1			1		1	
	Vallisneria spiralis			2	1				1				2	1		
5 - 6m	Ceratophyllum demersum	1					1					1	1			
6 - 7m	No plants															

MACROZOOBENTHOS OF SKADAR LAKE
REPORT FOR MONTENEGRO

Dr Ana Pavićević

August 2014

Contents

1. Introduction
(Purpose, objectives etc.)

This report focuses on the ecological assessment of Lake Skadar using benthic macroinvertebrates. The report's purpose is to review the current knowledge, current challenges, identifying the research needs in the use of benthic invertebrates as indicators of lake ecological status. The way forward on the realization of a lake holistic assessment tool based on benthic invertebrates is discussed in the light of the WFD requirements (Solimini et al. 2006).

The European Water Framework Directive (WFD) 2000/60/EC requires mandatory monitoring of the benthic macroinvertebrate communities and the ecological status/ potential assessment of freshwaters. Development of coherent sampling techniques to assess the benthic macroinvertebrates in lakes according to the requirements of the Water Framework Directive 2000/60/EC is a crucial challenge for each European country, particularly concerning local type. The WFD requires EU member states to assess, monitor and, where necessary, develop programmes of measures to improve the ecological quality status of all water bodies (lakes, rivers, coastal and transitional waters). These assessments should be based on standardised sampling or surveying methods for one or more biological quality elements (BQEs), including fish, macroinvertebrates, phytoplankton, diatoms and macrophytes, and habitats.

The present study investigations and a comparison of trends of benthic macroinvertebrate communities among three different habitats (littoral, sub littoral, profundal) of the lake in two different seasons in autumn (September 2013) and spring (April-May 2014).

Macroinvertebrates constitute a heterogeneous assemblage of animal taxa, many of which are sedentary, and some have relatively long life spans (life cycle of approximately one year or more) which makes them good indicators of localized conditions (Reice and Wohlenberg, 1993). Sensitive life stages respond quickly to stress while the overall community responds more slowly. Thus, the freshwater invertebrates can reflect both short and long-term shifts in water quality. Biological parameters integrate information over longer periods and better represent the responses of aquatic habitats, making biotic monitoring indices excellent tools for the sustainable management of water resources. Despite a wide range of adaptations, certain macroinvertebrate taxa can serve as indicators of environmental conditions and several groups of aquatic insects are among the organisms that are extensively used for monitoring aquatic ecosystems. The structure of the benthic communities in an aquatic ecosystem reflects its ecological conditions, including habitat heterogeneity (Heino et al., 2003) and water quality (Soldner et al., 2004). Some abiotic factors such as temperature, pH, electrical conductivity, dissolved oxygen in water column, granulometric composition and organic matter content in sediment, among others, determine the distribution of benthic macroinvertebrate communities (Allan, 1995). Benthic macroinvertebrate assemblages are made up of species that constitute a broad range of trophic levels and pollution tolerances, thus providing strong information for interpreting cumulative effects (Barbour et al. 1999). The use of biological indicators is more adequate to detect long-term changes in water quality, since aquatic organisms are adapted to specific environmental conditions. If these conditions change, some organisms may disappear (intolerant) and be replaced by others (tolerant). The first step in this type of biomonitoring is to find the ideal bioindicator whose presence, abundance and behavior reflects the effect of a stressor on biota (Bonada et al., 2006b). Some freshwater invertebrates are very sensitive to stresses produced by pollution, habitat modification, or severe natural events, while others are more tolerant. Benthic macroinvertebrates, such as annelids, crustaceans, mollusks, and adults and juveniles of insects, have biological and ecological characteristics that justify their wide use in biomonitoring studies. Distributions of bioindicator taxa are influenced by food availability, hydrological characteristics, nutrient supply, substrate type, predation pressure and natural or anthropogenic disturbances in addition to variation in water quality itself, which is what makes these biotic indices important tools for assessing the health of lakes.

Lake morphometry affects community structure of both macrophytes and macroinvertebrates (Duarte and Kalff, 1986). Generally, the benthic zone of lakes can be divided along the depth profile into the littoral, sub-littoral and profundal zones. The littoral zone is defined as the near-shore lake bottom areas where emerged macrophytes grow. The sub-littoral zone is defined as the bottom area covered by submerged macrophyte or algal vegetation. Often, empty shells of mollusks are accumulated at its lower end (littoriprofundal) and thus form a specific sediment type. The lake bottom area extending deeper is called profundal zone, which often consists of exposed fine sediment free of vegetation.

The identification of the type specific reference conditions, which should reflect high ecological status, for a specific lake type based on benthic invertebrates is complicated by the fact that the composition of benthic invertebrate communities exhibits natural variation due to season, lake depth, meso-scale habitat structure, and due to biotic effects (competition and predation). Therefore, reference states are to contain type-specific biological communities subjected to minor anthropogenic disturbance and minimally disturbed hydromorphological and chemical conditions. The reference state is, therefore, the benchmark from which all ecological classifications are to be made.

Freshwater environments are subjected to increasing degradation. In addition to the extensive range of natural stresses encountered by organisms in their habitats, human activities can generate other environmental stresses. Lakeshores have always been a preferential place for human settlement and various other human activities leading to shoreline development, and the discharge of wastewater on lakeshores represents a current threat to the ecological integrity of lakes worldwide. Moreover, the intensity of shoreline development is expected to increase in the future. Freshwater biodiversity provides a broad variety of valuable goods and services for human societies, some of them irreplaceable (Dudgeon et al., 2006), but human activities have always affected aquatic ecosystems. Globally, the biodiversity of freshwater ecosystems is rapidly deteriorating because of human activities. According to Dudgeon et al. (2006), there are five major threat categories to freshwater biodiversity: overexploitation, water pollution, flow modification, destruction or degradation of habitat and invasion by exotic species. Different groups of macroinvertebrates are excellent indicators of human impacts, especially contamination. Most of them have quite narrow ecological requirements and are very useful as bioindicators in determining the characteristics of aquatic environments. Improving our understanding of freshwater ecology is therefore very important not only because of its biological implications, but also because the proper management of freshwater is of practical interest to humanity.

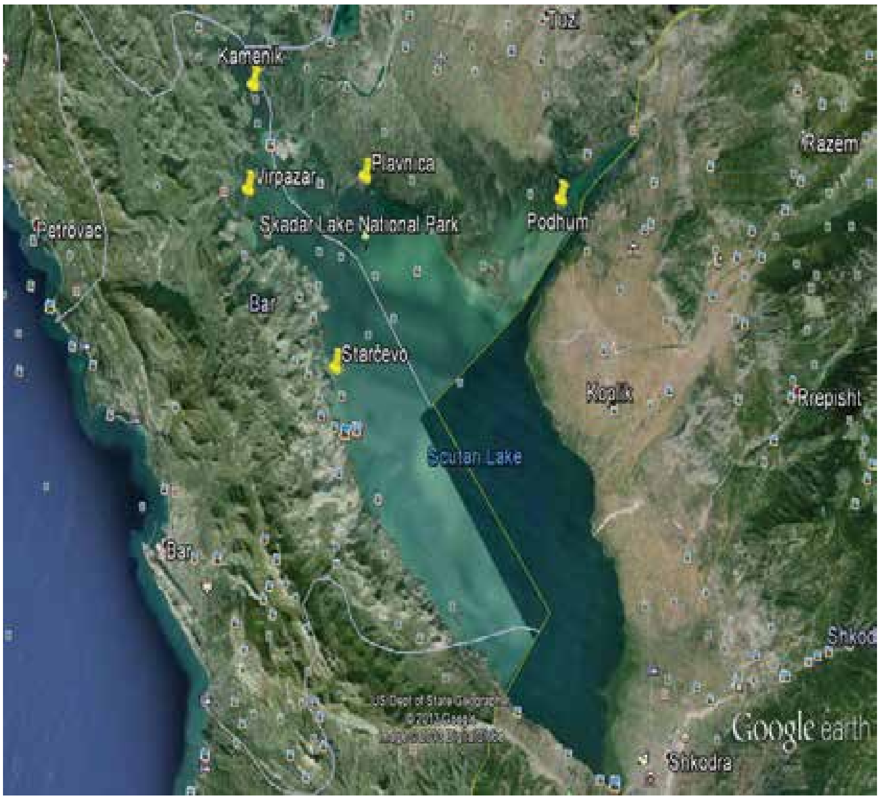
Current knowledge and examples of use are presented in the context of the required understanding needed to use benthic macroinvertebrates in lake assessment as required by Directive 2000/60/EC (see Table 1).

High status	Good status	Moderate status
The taxonomic composition and abundance correspond totally or nearly totally to undisturbed conditions. The ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels. The level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.	There are slight changes in the composition and abundance of invertebrate taxa compared to the type-specific communities. The ratio of disturbance sensitive taxa to insensitive taxa shows slight signs of alteration from type-specific levels. The level of diversity of invertebrate taxa shows slight signs of alteration from type-specific levels.	The composition and abundance of invertebrate taxa differ moderately from the type-specific conditions. Major taxonomic groups of the type-specific community are absent. The ratio of disturbance sensitive to insensitive taxa, and the level of diversity, are substantially lower than the type-specific level and significantly lower than for good status.

Table 1. Definitions for high, good and moderate ecological status in lakes for the biological element benthic invertebrate fauna (Directive 2000/60/EC).

2. Description of investigations
(Methods, standards, weather conditions, date of sampling etc.)

Study site
Sampling points at Skadar Lake (Montenegrin side):
- Virpazar
- Plavnica
- Starčevo



2.1 Starčevo

This characteristically pelagic sampling point is located at the western coast of the lake. It is located near to the extension of the Morača River, which is especially visible during torrential waters. Therefore, this corridor has specific hydrological and ecological characteristics, and Starčevo was selected as being representative of this zone. There are no local sources of pollution, or only negligible ones, but there is remote pollution from Morača River, and to some extent from the northern zone of the lake.

The first sampling campaign on the end of September 2013 included only the sub-littoral and profundal part of the lake:

- Point A - N 42 11 348; E 19 12 493 (profundal- which consists of exposed fine sediment (mud) free of vegetation; depth 4 m),
- Point B- N 42 11 460; E 19 11 668 (sub littoral- sandy-muddy bottom area covered by low density of submerged macrophytes, depth 1,5-2 m).

Second sampling company conducted in the beginning of May 2014 was extended on the littoral part of the lake:

- Point A - N 42^0 11^I $28,3^{II}$; E 019^0 12^I $06,9^{II}$ (profundal – consisting of exposed fine sediment, fine mud free of vegetation; depth 4 m),
- Point B- N 42^0 11^I $25,3^{II}$; E 019^0 12^I 04.0^{II} (sub littoral – sandy-muddy bottom area covered by low density of submerged macrophytes; depth 2,5m),

- Point C- N 42^0 11^I $22,6^{II}$; E 019^0 12^I $03,1^{II}$ (littoral – sandy-rocky bottom covered by submerged vegetation; depth 0-1.5m).

2.2. Plavnica

This sampling point is located downstream of the mouth of the river Plavnička. It belongs to the littoral part where the water is the shallowest of all measurement points, and represents the specific body of water on the lake. In addition to the hydrological characteristics, the influence of pollution of Podgorica and Zeta valley can be traced, where ground water moves toward the lake. It also comes close to monitor the impact of transport and catering facility in Plavnica (restaurant, hotel and marina).

The first sampling campaign on the end of September 2013 included the sub-littoral and profundal part of the lake:

- Point A - N 42 15 816 E 19 11 788 (depth 3.5m, bottom - muddy, covered by shells of dead mollusks);
- Point B - N 42 15 920 E 19 11 778 (depth 1.5 m, bottom - sandy-muddy with detritus and low density of macro-vegetation).

The second sampling campaign conducted in the beginning of May 2014 was extended to the littoral part of the lake:

- Point A - N 42^0 15^I $42,5^{II}$ E 019^0 12^I $02,3^{II}$ (depth 3.5m, bottom - muddy, covered by shells of dead mollusks);
- Point B - N 42^0 15^I $48,8^{II}$ E 019^0 12^I $01,5^{II}$ (depth 2.5 m, bottom - sandy-muddy with detritus and low density of macro-vegetation);
- Point C: N 42^0 15^I $57,7^{II}$ E 019^0 11^I $59,5^{II}$ (depth 1.5 m).

2.3. Virpazar

This water body is limited by the bridge Vranjina, cape Monastery Tapija and the western coast from

Radušato Virpazar. Therefore, the exchange of water with other water bodies is limited. Water depth is corresponding to littoral. There can be traced human impact by water utilities and other settlements of Virpazar, the canal of the Virpazarska River, the influence of water transport, which is very strong in this area, and the effect of the objects of the NP Skadar Lake and Plantaže 13 July at Vranjina.

The first sampling campaign at the end of September 2013 included the sub-littoral and profundal part of the lake:

- Point A: N 42 15 526 E 19 06 510 (depth 3.5m, bottom - muddy, covered by shells of dead mollusks);
- Point B: N 42 15 327 E 19 06 343 (depth 1.5m, bottom - muddy-sandy with detritus and low density of macro vegetation).

The second sampling campaign was conducted at the end of April 2014 at the littoral part of lake:

- Point A: N 42^{0} 15^{I} $26,0^{II}$ E 019^{0} 06^{I} $39,1^{II}$ (depth 4 m, bottom - muddy, covered by shells of dead mollusks);
- Point B: N 42^{0} 15^{I} $18,7^{II}$ E 019^{0} 06^{I} $35,1^{II}$ (depth 2.5m, bottom - muddy-sandy with detritus and low density of macro vegetation);
- Point C: N 42^{0} 15^{I} $14,3^{II}$ E 019^{0} 06^{I} $32,6^{II}$ (depth 0-1,5 m).

3. Data Analysis

BMWP and ASPT Biotic Indices

A Biological Monitoring Working Party (BMWP) score was used to deduce a biotic index of water quality, based on the macro-invertebrates identified. The macro-invertebrates were identified to family level and each family assigned a score depending upon their tolerance to nutrients, pollutants and low oxygen levels. The BMWP score is the sum of the individual family scores for a site (the higher the score, the better quality the water is as it includes more pollution intolerant families).. The BMWP scoring system is presented in Appendix and the evaluation criteria using BMWP scoring is presented in Table below.

Alternatively, also the Average Score Per Taxon (ASPT) score is calculated. The ASPT represents the average of the tolerance scores of all macro-invertebrate families found within community, and was calculated by dividing the BMWP score by the number of families represented in the sample (Friedrich et al., 1996). This score ranges from 0 and 10.

Standard BMWP scores and their related quality index.

BMWP score	Category	Interpretation
0-10	Very poor	Heavily polluted
11-40	Poor	Polluted or impacted
41-70	Moderate	Moderately impacted
71-100	Good	Clean but slightly impacted
>100	Very good	Unpolluted, unimpacted

Average Score Per Taxon (ASPT) representing an index of organic pollution

Average Score Per Taxa	Category
Over 5.4	Very good
4.81 – 5.4	Good
4.21 – 4.8	Moderate
3.61 – 4.2	Poor
3.6 or less	Very poor

As diversity indices are increasingly used to assess the well-being/health of the habitats, univariate measures included abundance (N), and number of benthic species (S), Shannon–Wiener (Shannon & Weaver 1963) diversity index, Margalef (1958) richness index (R=(S-1)/log2(N)R=(S-1)/log2(N)) and the Simpson index were calculated.

Relation between Shannon diversity index and Pollution level

Diversity level	Shannon diversity index	Pollution level
High	3.0-4.5	slight
moderate	2.0-3.0	light
less	1.0-2-0	moderate
Very less	0.0-1.0	Heavy pollution

Shannon index it's the most preferred index among the other diversity indices. The index values are between 0.0 – 5.0. Results are generally between 1.5 –3.5, and it exceeds 4.5 very rarely. The values above 3.0 indicate that the structure of habitat is stable and balanced; the values under 1.0 indicate that there are pollution and degradation of habitat structure.

And three diversity indices, obtained by using the formula of Margalef Diversity Indices (MDI), Simpson Diversity Indices (SDI)and of Shannon and Weaver Diversity Indices (SWDI) as detailed in Ludwig and Reynolds (1988) were evaluated.

Simpson (1949)		0,2-0,3	0,3-0,4	0,4 - 0,5	
Pielou (1966)		0,7-0,6	0,6-0,5	<0,5	
Shannon & Weaver (1963)		5,0 - 3,0	3,0 - 2,0	2,0 - 1,0	
Margalef (1958)	>8	8,0 - 6,5	6,5 - 4,8	4,8 - 3,5	< 3,5
Pantle & Buck (1955)	0,1 - 0,5	0,5 - 1,5	1,5 - 2,5	2,5 - 3,2	< 3,2
IBI	>4	>3	3	>2	<2
Water quality	Very good	Good	Moderate	Poor	Very poor

Environmental quality assessment, based on the diversity indices

Margalef water quality index values greater than 3.0 indicate clean conditions, values less than 1.0 indicate severe pollution and intermediate values indicate moderate pollution (Lenat et.al, 1980). This index shows variation depending on number of species, so that the number of individuals is less important for calculation.

Lenat, D.R., Smock, L.A. and penrose, D.L. (1980). Use of benthic macroinvertebrate as indicators of Environmental Quality. In: Biological monitoring for environmental effects. Douglass, L.W. (ed.). Published by Lexington books. Toronto.pp. 97-114.

Tentative Quality Ratings: Revised 2004

Water Quality	Taxa Richness	EPT Taxa Richness	MBI
Excellent	>= 14	>= 5	<= 4.35
Good	12 - 13	4	4.36 - 5.00
Fair	9 - 11	3	5.01 - 5.70
Poor	7 - 8	2	5.71 - 6.25
Very Poor	<= 6	0 - 1	>= 6.26

4. Collection & Identification of Macroinvertebrates

Sampling seasons:

September 2013; weather: cloudy with wind, transparency of water very low

26 April 2014; weather: Rainy with wind; transparency of water very low

02 May 2014; weather: Cloudy with wind

Sampling methods: Standard limnological methods ISO: EN 27828:1994 (AQEM/STAR), Multihabitat transect method.

Main equipment: Van Veen grab (250cm^2); D-shape net (400µm mesh size); Macrobenthic sieves: (0.5 mm mesh size); Binocular stereomicroscope (20-100X)

Main consumables: 1000 ml plastic containers for field samples; 30- 100 ml plastic containers for sorted samples; 70% ethanol; 4% formalin.

Benthic macroinvertebrates were collected at three study sites. Sampling was carried out in autumn 2013 and spring 2014. At each study site and sampling occasion, 18 samples were taken from different depths (littoral, sub-littoral and profundal). From each sampling point, two quantitative samples were taken using Van Veen grab. The samples were fixed in 4% formalin solution. In the laboratory, the samples were washed, sieved and all macroinvertebrates sorted under a stereomicroscope. Specimens of macroinvertebrates were identified to the species level. Because of the low number of captured specimens, there was no need to predefine a minimum number of individuals to be sorted per sample.

The WFD requires the collection of data informing on the communities' taxonomic composition, abundance, diversity and sensitive taxa. Indices assessed in this study for the determination of the ecological status were the following: Total number of taxa; relative abundance of taxa; dominance; diversity indices such as Shannon & Wiener index (H'), Simpson and Margalef index and status indices such as: Macroinvertebrate Biotic Index (MBI), Biological Monitoring Working Party score (BMWP) and Average Score Per Taxa (ASPT).

The taxa were also classified according to functional feeding groups (FFG) which categorize invertebrates based on their feeding mode. In contrast to other biometrics that measure the structure of the invertebrate community, FFG analysis measures its functioning. By using these functional feeding group definitions, the invertebrates present in the community indicate the type and relative amounts of different food sources utilized. They also describe ongoing processes within the lake. Changes in the proportions of these functional feeding groups can indicate stressful conditions, as well as changes of the resource base.

Species diversity indices such as Shannon-Wiener, Simpson's, and Margaleg are computed to understand biotic communities (Chakrabarty and Das, 2006). The Shannon-Wiener diversity index relates to species relative abundance, Simpson's diversity index weighs towards the abundance of the most common species. The Margalef index has good discriminating ability and is sensitive to sample size. It is a measure of the number of species present for a given number of individuals.

According the fact that this study can serve as the initial phase of monitoring establishment of water quality of Lake Skadar, it was decided that during the second sampling campaign (spring season), the sampling should cover different depth zones of the lake. After the initial phase, the most representative sampling sites can be selected. The sites should reflect the situation prevailing in a significant part of the lake and influencing the bottom fauna to a considerable extent.

In case of lakes, appropriate metrics for the optimal interpretation of ecological status are not widely accepted yet. Until now, no significant steps have been taken towards inter-calibration for lakes of different geographical intercalibration groups (GIGs).

Van Veen grab

5. Results and discussion

There is quite some literature, mostly research papers and reports, on the quality of water and sediments at Lake Shkoder and in its tributary rivers. However, the literature suggests that analyses of water quality of the lake before 1990 are fragmentary. The most comprehensive analysis of the lake water and its tributaries is presented in the book "The Biota and Limnology of Lake Skadar" (Karaman, G.S. & Alfred M. Beeton, 1981) based on the data collected in the period 1974-1978. It can be considered as a basis for evaluation and comparison, as the lake until then had hardly been affected by pollutants such as agricultural fertilizers, chemical detergents; municipal and industrial wastes (Petrovic, 1981; p.68 and p.97). Various reports on Shkoder basin suggest that during the past decades the basin has experienced significant pollution of its water bodies and the surrounding area, causing losses in aquatic biodiversity and a threat to fisheries, public health and tourism. Most important are industrial pollution and untreated wastewater discharges from the cities and towns along River Morača, the main tributary of the lake, which brings most pollutants into the lake.

The annex V of the WFD specifically outlines benthic invertebrate fauna composition and abundance, the ratio of sensitive taxa to insensitive taxa and the diversity of invertebrate communities as criteria that need to be defined for type-specific ecological assessments of lakes. According to Cummins et al. (2005), the proportion of macroinvertebrate functional feeding groups can be used as a measure of the trophic state of an aquatic ecosystem.

Macroinvertebrates were sampled in autumn (in September 2013) and spring (in April-May 2014) at each of the three sampling stations (Starčevo, Virpazar and Plavnica) from different water depths (littoral, sub-littoral and profundal part). Assemblage structures were similar between assessment sites in both surveys (Table 2A, 2B) and there were no considerable differences in community composition among seasons, nor in major macroinvertebrate or functional feeding groups (see Appendices).

The Percent Contribution of Dominant Family or percent dominance (DF) equals the abundance of the numerically dominant family relative to the total number of organisms in the sample. This index indicates the present state of the community balance at the family level. For example, a community dominated by relatively few families would have a high DF value, thus indicating the community is under the influence of environmental stress (Plafkin et al., 1989). Numerically dominant families were similar across investigated sites; Oligochaeta and Diptera followed by Bivalvia, contributed the greatest number of individuals and Crustacea, Odonata and Coleoptera, the least (Table 3A, 3B). The structure of benthic macroinvertebrates varied greatly among sites in both seasons, ranging from 3 to 10 families. Overall, the number of individuals in both sampling seasons was highest in Starčevo while the taxonomic diversity was highest at Virpazar and lowest at Plavnica. Species richness was highest in the order of Diptera in both seasons.

In spring, littoral species richness was significantly higher than profundal species richness, but was not significantly between sub littoral and profundal, while in autumn, species richness did not differ significantly between the sub littoral and profundal part of the lake except for the sampling point Starčevo.

During the first sampling campaign in autumn 2013, 214 specimens were collected. These belonged to 7 taxonomic groups. The most abundant taxonomic group were Bivalvia (family Dreisseniidae: 63.1%) and Oligochaeta (Tubificidae: 17.3%), representing about 80% of the total benthic macroinvertebrates, with several groups comprising less than 1% of the total (Table 2A). During the second sampling campaign in spring 2014, 342 individuals were collected. These belonged to 8 major taxonomic groups. The most abundant taxonomic group were Oligochaeta (41.5%) and Diptera (33.6%) at all three sites, followed by Bivalvia (20.8%) and several groups comprising less than 1% (Trichoptera, Odonata, Coleoptera) of the total benthic macroinvertebrates (Table 3B).

The number of specimens found during the spring sampling campaign was slightly higher than during the preceding autumn sampling campaign. Another crucial factor might be the limited number of samples concerning the total length of Lake Skadar as well as the position of the sampling sites.

In the littoral zone of lakes, the mosaic of habitat types creates a high spatial heterogeneity and usually supports larger and more diverse populations of benthic invertebrates than do the sub-littoral and profundal habitats, which is associated with a high diversity in food resources. Therefore, littoral macroinvertebrate species richness, abundance and biomass are higher than in the sub-littoral or profundal zones, which was confirmed in our survey. The vegetation and substrate heterogeneity of the littoral habitat provide an abundance of microhabitats occupied by a varied fauna, which in turn enhances invertebrate production. The littoral habitat is also highly variable due to seasonal influences, land use patterns, riparian variation, and direct climatic effects producing high-energy areas.

The sub-littoral habitat, below the area of dense macrophyte beds, lacks the heterogeneity of the littoral habitat. However, it is also less subjected to littoral habitat variables and influences. The sub-littoral habitat is rarely exposed to severe hypoxia but might also lack the sensitivity to toxic effects that is found in the profundal habitat. The profundal habitat, in the hypolimnium of stratified lakes, is more homogeneous due to a lack of habitat and food heterogeneity, and hypoxia and anoxia in moderately to highly productive lakes are common.

According to literature data, the profundal habitat is usually dominated by three main groups of benthic organisms, including chironomids larvae, oligochaete worms, and phantom midge larvae (*Chaoborus*). Many species of chironomids and tubificid oligochaetes are tolerant to low dissolved oxygen, such that these become the dominant profundal invertebrates in lakes with hypoxic hypolimnia. As hypoxia becomes more severe, tubificids can become dominant over chironomids. In case of prolonged anoxia, the profundal assemblage might disappear entirely. Consequently, the assemblage of organisms living in the profundal zone can provide an indication of past and current disturbances and may be used in the assessment of the ecological conditions of a given lake (Brinkhurst 1974; Rosenberg & Resh 1993).

In both sampling campaigns, results for benthic macroinvertebrate communities showed less taxonomical diversity than expected. There were few differences in species composition and structure of benthic communities between sampling campaigns and substrates (mud and gravel/stones). The profundal zone showed the lowest taxonomic diversity and was dominated by only a few taxa that are very resistant to low water oxygenation. Assemblages of organisms living in the profundal zone were Oligochaeta, Chironomidae (Diptera) and Bivalvia (*Dreissena polymorpha*). In autumn at the profundal zone at Virpazar, chironomids dominated over oligochaetes, at Starčevo chironomids were slightly more abundant than oligochaetes while oligochaetes were entirely absent at Plavnica where only two individuals of species *Chironomus pulminosus* were found. In spring at the profundal zone of Starčevo, oligochaetes (Tubificida**e**) were more abundant than chironomids. At Virpazar, the reverse was true while at Plavnica, both taxa were about equally represented. Considering lake typology and the predominance in both sampling campaigns of profundal Chironomidae, notably the by far most dominant species *Chironomus plumosus*, the lake can be classified as belonging to the hyper-eutrophic type.

The low diversity of macroinvertebrates in the profundal and sub-littoral zones seems to be related to the low diversity of habitats. Habitat diversity is the main driver of species diversity in benthic communities since different species require different habitats. Furthermore, the abundance and diversity of macroinvertebrates decreases with increasing depth. The low abundance of insect groups in our study can be explained by the fact that their presence in the benthos varies according to many factors (acting alone or in combination) such as the distance from the littoral zone, depth, oxygenation and water quality, predation by certain groups, sediment composition, and life histories.

Figure1A. Structure of autumn macrozoobenthic community

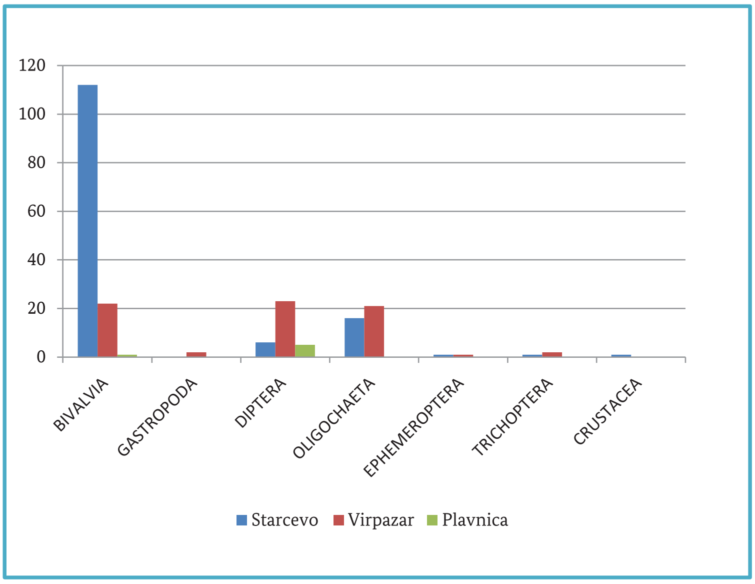
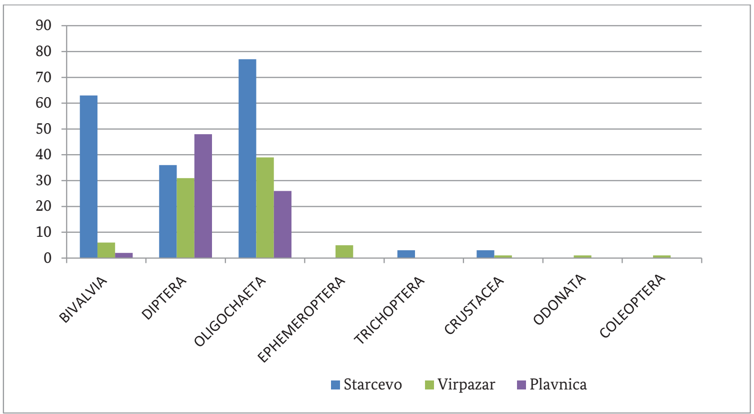


Figure1B. Structure of spring macrozoobenthic community



Our research has confirmed once again the fact that macrophyte cover generally supports greater diversity and abundance of invertebrates than open silty areas or those dominated by gravel and stones (Watkins et al., 1983) and removal of submerged vegetation generally reduces macroinvertebrate taxa richness (Rabe & Gibson, 1984; Tolonen et al., 2003).

In the profundal zone, oligochaetes and chironomids are considered the most useful indicators of oxygen conditions and trophic status (Solimini et al 2006). Therefore, species composition changes with changing lake trophic status. Oligochaete and chironomid species are complementary indicators because of differences in their autecological traits. Chironomid larvae are more mobile and can migrate at different depths, being less dependent on the quality of the sediment than the oligochaetes. Moreover, chironomid larvae feed on "fresh" detritus deposited on the surface of sediments, whereas oligochaetes feed on bacteria associated with organic matter in an advanced state of decomposition. Therefore, chironomids are preadapted to react more rapidly than oligochaetes to changes in environmental conditions, and they are expected to react more quickly to the improvement of water quality in lakes (Dinsmore and Prepas, 1997; Lang and Lods-Crozet, 1997). Recently, research of freshwater Oligochaeta as indicators in water quality assessment (Sundić and Radujković, 2012) has shown that the lake is at the mesotrophic-eutrophic level, depending on sampling location and period. The values of saprobic index were ranging from β-α-mesosaprobic, which indicates highly or excessively polluted waters.

Sensitive taxa may be rare and detection can vary with sampling effort. Low impacted sites tend to have greater taxa richness and more rare species than impacted ones (Doberstein et al., 2000; Chase and Liebold, 2002). For this reason, a number of authors (Lyons et al., 1995; Cao et al., 1998) have argued that rare species are critically important indicators of ecosystem health. Benthic communities with a large proportion of pollution-sensitive organisms usually are associated with water of relatively high quality. In contrast, benthic communities in water badly polluted with organic waste usually contain large proportions of pollution-tolerant organisms. In view of this, we conclude that the absence or low number of sensitive (rare) species at the sampling sites investigated in the present study is a clear indication that Skadar Lake is suffering considerably from pollution.

The use of invertebrate sample analysis is a useful tool for calculating the overall health of a lake, as certain species of invertebrates are more intolerant than others to inorganic and organic pollution, which may or may not be present within a water body (Tierney 2013). Benthic macroinvertebrate species are differentially sensitive to many biotic and abiotic factors in their environment. Consequently, macroinvertebrate community structure has commonly been used as an indicator of the condition of an aquatic system (Rosenberg and Resh, 1993). Biotic index systems have been developed which give numerical scores to specific "indicator" organisms at a particular taxonomic level. Such organisms have specific requirements in terms of physical and chemical conditions. Changes in presence/absence, numbers, morphology, physiology or behaviour of these organisms can indicate that the physical and/or chemical conditions are outside their preferred limits (Rosenberg and Resh, 1993). Presence of numerous families of highly tolerant organisms usually indicates poor water quality (Hynes, 1998). The pollution tolerance of the organisms present varies from the considered sensitive to environmental stress (Ephemeroptera, Trichoptera) to taxa considered as very tolerant of organic elements (Oligochaeta, Gastropoda and Diptera-Chironomidae).

One of the aims of this survey was to use BMWP (Biological Monitoring Working Party) and ASPT (Average Score per Taxon) scoring systems to assess the general health of Lake Skadar from the available data obtained through an initial baseline survey, and also to provide a base line of aquatic macro-invertebrates present in the spring and autumn seasons. The BMWP score is based on the principle that different aquatic invertebrates have different tolerances to nutrients, pollutants and low oxygen levels. With this information, families of invertebrates have been assigned individual biotic scores such as the BMWP score. The calculation of biotic indices to assess water quality is an established method. The scores were primarily developed for identifying organic pollution but they are also widely used as indicators of the general health of a water body. A high BMWP score indicates good water quality. Naturally species-rich sites have higher scores than naturally poor sites even if the physico-chemical water quality is the same. Therefore, to overcome this dilemma, an ASPT can be derived by dividing the BMWP score by the number of BMWP families in the sample, yielding an index of organic pollution (Wright et al, 2000) that does not depend on family richness.

The list of aquatic invertebrates recorded in autumn gave a BMWP score of 26.5 and 22.1 for Starčevo and Virpazar, respectively, which indicates a polluted and impacted site, while at Plavnica the BMWP score was only 8.9, indicating a heavily polluted site (Table 4A). An ASPT score of 3.3 was obtained for Starčevo which has a category designation of very poor water quality (Table 4A), for Virpazar and Plavnica 4.4 and 4. 5 respectively, which has a category designation of moderate (fair) water quality. The scores for the taxa via the ASPT system are more informative than the BMWP score and relate to the average of the tolerance scores of all macro-invertebrate families found in the samples. The water quality of Lake Skadar based on BMWP and ASPT scores indicates substantial pollution.

The BMWP scores recorded in spring were 25.5 for Starčevo, 29.6 for Virpazar and 12.4 for Plavnica, which largely confirmed the results from the autumn assessment. An ASPT score of 3.6 and 4.1 for Starčevo and Plavnica, respectively, indicated poor water quality and the score of 3.0 for Virpazar very poor water quality.

A Macroinvertebrate Biotic Index (MBI) is an established scientific procedure that is used to monitor pollution levels of water bodies worldwide. According to our results from all three sampling sites and both seasons, the MBI scores indicated very poor water quality and a severe degree of organic pollution of the lake.

Taxa have different pollution tolerances and the level of tolerance is reflected in the predominance of certain taxa in specific water quality conditions. Oligochaeta in high numbers indicate very poor water quality. In our autumn investigation, they were recorded at two sampling sites (Starčevo and Virpazar) but were absent from Plaice, while in spring they were represented at all three sites. Mussels (Bivalvia) are known to be sensitive elements of benthic communities, reacting to siltation and low dissolved oxygen. In both seasons in the present study, Bivalvia (mainly *Dreissena polymorpha*) were one of the dominant groups at Starčevo and Virpazar but were absent from Plavnica. However, in spring the family Unionidae (Bivalvia) was found at Plavnica. This family is moderately tolerant of organic enrichment. The class Gastropoda can indicate nutrient-enriched conditions and poor water quality. In both seasons, live gastropods were found only in Virpazar (*Viviparus acerosus*) while in autumn only empty shells were registered. We found a huge amount of empty shells of other gastropods: Valvatidae, Hydrobiidae (*Pyrgula annulata*, *Radomaniola* sp.), Ancylidae (*Ancylus fluviatilis)*, Neritidae (*Teodoxus fluviatilis)*, Lymnaeidae, Bithyniidae and others. Even though species of Trichoptera show a large range of pollution tolerance, the trichopteran families are generally indicators of oligotrophic conditions and usually live in well-oxygenated waters (Pérez, 1988). In this order, members of some families are highly sensitive to habitat degradation, while others can survive under certain levels of pollution such as the family Polycentropodidae found during our research in both seasons. Ephemeropteran families vary in their tolerance to pollution but generally live in clean water. At Starčevo in autumn and Virpazar in spring, we found *Caenis horaria* which has a tolerance value of 6, indicating moderately clean water (This species can tolerate some degradation of water quality due to organic input). Among the crustaceans, the gammarid *Asellus* is more tolerant of organic pollution than *Gammarus*.

Dipterans were more common and comprised on average 15.8% of the autumn sample and 33.6% of the spring sample. The dipteran community consisted primarily of chironomids (*Chironomus plumose*). Unlike most insects, some chironomids contain haemoglobin, which enables them to survive in low oxygenated areas. Because of this, their presence in high numbers is often one of the first signs of pollution and organic enrichment. This was the most common taxon within the samples (present at all three sites). According to literature data, the dominance of pollution-tolerant oligochaetes and dipterans indicates a degradation of water quality. Tables 3A/3B (see Appendix) show that Diptera were dominant at two sites in autumn (Virpazar and Plavnica) and one site in spring (Plavnica).

Taxonomic richness was different between sampling stations. It indicates the health of communities through diversity and increases with increasing habitat diversity, suitability and water quality (Plafkin et al., 1989). The healthier the community is, the greater the number of taxa found within that community. In the present study, the low number of taxa at Virpazar and Plavnica in autumn indicated very poor water quality and severe organic pollution. Conditions were slightly better at Starčevo, indicating poor water quality and very substantial pollution. Results in spring showed very poor water quality and severe organic pollution at both Starčevo and Plavnica, and poor water quality and very substantial pollution at Virpazar.

Three diversity indices were used in the present study, the Shannon-Wiener, Simpson and Margalef indices (the higher the value of the respective index the greater the diversity). The usefulness of these indices for assessing water quality is based on the fact that biodiversity is usually higher in clean water and that benthic communities of clean water bodies contain many species of relatively equal numbers of individuals per species (Wilhm and Dorris, 1968). In contrast, polluted waters are known to have low diversity because many pollution-sensitive species disappear and only a few pollution-tolerant organisms flourish in the absence of competition and the presence of an abundant food supply. According to Wilhm (1970), "H usually varies between three and four in clean-water stream areas and is usually less than one in polluted-stream areas." In general, low values of diversity are associated with eutrophic lakes. In the present study in autumn, Shannon diversity was highest at Virpazar, and lowest at Plavnica (1.9 and 0.6, respectively), while Simpson diversity was highest at Plavnica and lowest at Virpazar. Species richness (Margalegf) was lowest at Plavnica (1.3) and highest at Starčevo (3.7). Low species diversity indicates: relatively few successful species in the habitat; the environment is quite stressful with relatively few ecological niches and only a few organisms are well adapted to that environment; food webs which are relatively simple; change in the environment would probably have quite serious effects. From Table 5A below it is clear that in autumn 2013 all indices indicate poor or very poor water quality, with slightly better conditions at Starčevo where only one of the three indicators points to very poor conditions.

During the spring survey, Shannon diversity was highest at Virpazar, and lowest at Plavnica (1.8 and 1.1, respectively), while Simpson's diversity was highest in Virpazar and similar between Starčevo and Plavnica. The Margalef index was highest in Virpazar (2.7) and lowest in Plavnica (0.9). The results largely confirmed the results from the autumn survey with the exception of the Margalef index, which was more than two times higher in autumn than in spring. This index shows variation depending on number of species, so that the number of individuals is less important for calculation.

Clear seasonal changes in community structure can be observed which are primarily due to the life cycles of aquatic insects but may be influenced by seasonal changes in habitat conditions, too. As most univoltine insects emerge in summer, these are hardly present in summer samples of benthic invertebrates, and the early larval stages present in autumnal samples can often not be determined taxonomically. Some species have only one generation every year, others several generations and a few take several years (2-3 years) to complete one generation (Chironomidae). On the other hand, emergence of adults from aquatic larvae may occur during the entire year.

The nature of sediments is an important factor in shaping benthic communities, both in structural and functional composition. Sediments with high percentage of sand fractions have well distributed interstitial spaces, which favour oxygenation of sediment. All three investigated sites in our survey have similar substrates (mud, clay, gravel), providing poor habitats for benthic macroinvertebrates. These habitats normally support organisms such as Oligochaeta, certain chironomids, and pulmonate snails that are highly tolerant of organic enrichment.

According to CUMMINS et al. (2005), the proportions of macroinvertebrate functional feeding groups can be used as a measure of the trophic state of aquatic ecosystems. Feeding strategies are typical traits reflecting the adaptation of species to environmental conditions. Using these functional feeding group definitions, the invertebrates present in the community indicate the type and relative amounts of different food sources utilized. They also describe ongoing processes within the lake. Changes in the proportions of these functional feeding groups can indicate stressful conditions, as well as changes to the resource base.

The specimens found in the lake were classified into five functional trophic groups (Table 2), defined by their feeding strategy as collector-gatherers, predators, collector-filterers, shredders, and scrapers (CUMMINS et al., 2005). The collector-filterers were the most abundant trophic group, comprising 63.38% of the all macroinvertebrates specimens from all three sites in the autumn survey. This can be explained because of the abundance of resources produced by the rapid degradation of allochthonous organic matter. The second most abundant were the collector- gatherers, with 32.86% of the total. The predators comprised (2.34%), followed by scrapers (0.93%) and shredders (0.47%). The presence or absence of certain feeding groups such as grazers and predators also reflects water quality. The presence of four of the five feeding groups (Starčevo, Virpazar) suggests a balanced benthic community within the lake. Plavnica is the exception with only collector-gatherers and collector-filterers being present (Table 2) . Dominance of a particular feeding group corresponds to the abundance of a particular food source, which reflects a specific type of impact on the community (Plafkin et al., 1989). Collector-filterers were the dominant FFG at Starčevo, which can be explained by the predominance of Bivalvia, while the dominance of collector-gatherers at Virpazar and Plavnica was due to the dominance of Diptera. Abundant supply of suspended fine particulate organic matter support large populations of filtering collectors. The number of predators recorded in this study was low, they were even completely absent at Plavnica. This was expected since specialized feeders are more sensitive and usually more abundant in healthy lakes. Generalists, such as collector-filterers, which were well represented in this study, have a broader range of acceptable food materials than specialists (Meritt et al., 2002) and thus are more tolerant to pollution that might alter the availability of certain food. Their wide-spread distribution in the study area may be indicative of the perturbed status of the lake.

A general comparison of functional feeding groups of benthic macroinvertebrates in both seasons suggests a similar structure across investigated sites, with four major FFGs recorded as well in the spring survey, including predators, shredders, collector-gatherers (and collector filterers. Analysing the functional composition of the assemblage, it was found that collector gatherers were the most abundant FFG (Table 6B) accounting for 73,97% of the animals recorded in spring survey. Collector-filterers ranked second in total density of individuals and constituted 20.76% of the total population, while predators and shredders accounted for 3.5% and 1.46 % of the total population, respectively. Diversity of macroinvertebrates with the functional feeding groups is listed in Table 6B.

Factors affecting water quality of the lake can be classified as natural and human made. Human intervention is the most important factor influencing water quality, and it must be controlled. Profundal and littoral benthic communities may respond differently to various types of anthropogenic stresses. Littoral communities respond more rapidly to the anthropogenic impacts compared to profundal invertebrate communities. In contrast, profundal communities may reflect long-term changes in environmental conditions of a lake. Consequently, the assemblage of organisms living in the profundal zone can provide an indication of past and current disturbances and may be used in the assessment of the ecological conditions of a given lake (Brinkhurst 1974; Rosenberg & Resh 1993).

The relatively shallow depth of Skadar Lake (less than 10 m) and oscillations in water level lead to large oscillations in water temperature during the year. As a result, there is no clear depth stratification of macroinvertebrate habitats and no clear separation into littoral, sub-littoral and profundal zones. In shallow lakes, ecological thresholds in response to nutrient conditions are much more difficult to define due to feedback-effects of macrophytes and fish on lake community structure.

The riparian zone is also an important factor for the perfect functioning of the lake ecosystem, because it provides food and shelter for the aquatic biota, modifies the microclimate (light, temperature and humidity), contributes organic matter to the lake (from large, woody debris to small particulate matter) etc. The importance of vegetation (riparian and macrophytes) as a source of allochtonous and autochthonous organic matter and as a structuring factor for macroinvertebrate communities has been well established (Burton et al. 2002; Kelly and Hawes 2005; Marchese et al. 2005; Poi de Neiff et al. 2006). Riparian vegetation of all three sites investigated was well represented (willow formations, Montenegrin forest of ash, oak and alder, and southern riparian galleries and thickets). *Trapa longicarpa* ssp. *scutariensis* is widespread and together with white and yellow water lily represents the majority of the floating vegetation. Submerged vegetation such as *Potamogeton lucens, Najas* sp., *Ceratophyllum submersum, Myriophyllum spicatum* and *Valisneria spiralis* was noted up to 3.5 m depth.

Ancient lakes such as Lake Skadar (Glöer and Pešić, 2008a) are among the most vulnerable and threatened ecosystems (Lévêque et al. 2005) and their faunas are frequently under extreme anthropogenic pressure (Coulter et al. 2006). The small range of endemic species living at the Skadar Lake system together with ever increasing human pressure make its fauna particularly vulnerable. Investigation of freshwater snail (Glöer and Pešić 2008) state that this becomes even more important in light of ongoing eutrophication, pollution and sand and gravel exploration at the lake and its basin. Recently, research of phytoplankton community and chlorophyll-based trophic state indices (Rakočević, Nedović and Hollert 2005) showed that the lake is on a beta-meso-saprobic level of saprobity, which means moderately polluted with organic compounds. The eutrophic level in the lake was confirmed during August by the investigations of phytoplancton (Rakočević, 2006), when the contidition of the extreme eutrophy was determined.

Parameter	Sampling site and season (Autumn 2013/spring 2014)					
	STARCEVO		VIRPAZAR		PLAVNICA	
	Autumn	Spring	Autumn	Spring	Autumn	Spring
MBI	9.1	9.0	10.0	8.4	10.0	10.0
BMWP	26,5	25.5	22.1	29.6	8.9	12.4
ASPT	3.3	3.6	4.4	3.0	4,4	4,1
Shannon–Wiener	1.1	1.4	1.9	1.8	0.6	1.1
Simpson	0.7	0.7	0.3	0.7	0.7	0.6
Margalef	3.7	1.5	2.7	2.7	1.3	0.9
Feeding groups (%)	%					
Scrapers	0.2		2.8			
Collector-filters	81.7	34.6	31.4	7.1	16.7	2.6
Collector-gatherers	15.3	61.5	62.8	85.7	83.3	90.7
Predators	2.1	2.1	2.8	3.6		1.3
Shredders	0.7	1.6		2.4		

Table 2. Summary of results from macrozoobenthos investigations conducted at Lake Skadar in 2013 and 2014. Water quality/category: yellow – moderate; orange – poor; red – very poor

6. Conclusion

Macroinvertebrate composition, abundance and distribution are influenced by water quality. The main objective of this study was to determine the qualitative and quantitative structure of the macrozoobenthos in Lake Skadar, to use the results as an indicator of the Lake's ecological state. The differences between taxonomic composition and structure of macroinvertebrate assemblages were not notable between two seasons nor in major (dominant) macroinvertebrate or functional feeding groups.

The bottom of Lake Skadar comprises mostly muddy deposits that are accompanied by detritus and mollusk shell accumulations in deeper zones. Biological analyses pointed to the presence of 8 benthic fauna taxa at the bottom of Lake Skadar, comprising mostly Oligochaeta (Tubificidae) and Diptera larvae (Chironomidae, Ceratopogonidae and Tipulidae) which are highly resistant to unfavourable habitat conditions. In the shallow (littoral) zone, class belonging to the Oligochaeta (Tubificidae), Bivalvia (*Dreissena polymorpha*), Insecta (Trichoptera, Diptera, Coleoptera, Ephemeroptera, and Odonata larvae) and Crustacea were found. There were 3 shared taxa throughout the entire water body (deep and shallow zones). Shallow zones were characterised by a higher density of benthic fauna than the deeper zones. The euconstant species in both investigated seasons, regardless of location, were Oligochaeta and Diptera larvae, followed by Bivalvia. The remaining taxa were classified as accidental species. The scant occurrence of insect larvae (Ephemeroptera, Trichoptera, Odonata and Coleoptera) and crustaceans, which are predominantly accidental species, indicates that the lake offers poor habitat conditions.

Low taxonomic diversity and dominance of certain groups of macrozoobenthos (Diptera and Oligochaeta) and the possible replacement of pollutant-sensitive species with pollution-tolerant ones seem to be the result of initial pollution. During both investigation periods, a high abundance and dominance of Oligochaeta and Chironomidae (indicators of beta- and alpha-mesosaprobity) particularly at Plavnica indicated low water quality. The presence of Ephemeroptera, Trichoptera and Gammaridae at Starčevo and Virpazar indicates that this part of the lake was much less polluted than Plavnica. Overall, the benthic fauna reflects a tendency of eutrophication, nutrient-enrichment and organic pollution.

All biotic indices used in the present study lead to the conclusion that the ecological status of Lake Skadar is poor or even very poor, with Plavnica being in the worst condition.

Ideally, sampling should be undertaken at least twice a year as the phenology of different aquatic invertebrates can vary during the season. Results of the present investigation provided no clear evidence of seasonality since the number of species and the total number of individuals were very similar. The overall low number of specimens sampled during the two campaigns may have been the result of adverse weather conditions, even though the exact reason could not be fully elucidated.

Due to on-going threats to the aquatic ecosystems of Lake Skadar, a better understanding of lake biodiversity is needed to devise specific conservation measures. Lake Skadar in recent years is under intense anthropogenic pressure resulting from economic development, urbanization, deforestation and other factors. In Montenegro, there is no monitoring of environmental health of aquatic environments in place.

We expect to continue to monitor the ecological status of the lake and extend the sampling to a few more sites, particularly to the severely disturbed areas of Lake Skadar. This study provides the basis for continued monitoring based on existing assessment models but also opens up the option to develop new models specifically adapted to Lake Skadar.

Therefore, water management and water protection need to include knowledge from bio-monitoring for sustainable management decisions. Biomonitoring using biological indicators makes it possible to monitor the frequency and duration of impacts and provides data on species loss, magnitude of impact and baselines for measures designed for recovery.

The assessment of the ecological status of lakes and other water bodies is a long learning process. As the biological elements are not often included in routine monitoring, experience is needed to be able to use these elements properly. These statements clearly indicate that the implementation of the WFD is not an easy and rapid issue to solve but a challenging and time-consuming learning process for years to come. Results from this project, for sure, will serve as a recommendation for expanding and modifying the monitoring programme according to the WFD in the future.

7. Appendices

TAXON	STARCEVO		VIRPAZAR		PLAVNICA		FFG
	A	B	A	B	A	B	
ANNELIDA							
OLIGOCHAETA							
Tubificidae	+	+	+	+			c-g
MOLLUSCA							
GASTROPODA							
Viviparus acerosus				+			scr
BIVALVIA							
Unionidae							
Anadonta sp.						+	
Dreisseniidae							
Dreissena polymorpha	+	+	+	+			c-f
Sphaeriidae							
Pisidium casertanum	+						c-f
INSECTA							
DIPTERA							
Chironomidae							
Subfamily Tanypodinae							
Tanypus sp.	+						prd
Subfamily Chironominae							
Chironomus pulmosus	+	+	+	+	+	+	c-g

Subfamily Orthocladiinae				+			c-g
Ceratopogonidae							
Bezzia sp.		+					prd
EPHEMEROPTERA							
Caenidae							
Caenis sp.		+					c-g
TRICHOPTERA							
Polycentropodidae							
Phylocentropus sp.		+		+			prd
Decapoda							shr
Palaemonidae							
Palaemonetes antennarius		+					

Table 3 A. Structure of autumn macrozoobenthic community. A-profundal part of lake, B-sub littoral part. FFG-functional feeding group

TAXON	STARČEVO			VIRPAZAR			PLAVNICA			FFG
	A	B	C	A	B	C	A	B	C	
ANNELIDA										
OLIGOCHAETA										
Tubificidae	47	18	12	3	19	17	10	12	4	c-g
MOLLUSCA										
BIVALVIA										
Unionidae										
Anadonta	1						1		1	c-f
Dreisseniidae										
Dreissena polymorpha	14	18	30		2	4				c-f
INSECTA										
DIPTERA										
f. Chironomidae										
Subfamily Tanypodinae										
Tanypus sp.									1	prd
Subfamily Chironominae										
t. Chironomini		2	12	3		7				c-g
Chironomus pulmosus	8		6	6			10	11	14	c-g
t. Tanytarsini			7			2				c-g
Subfamily Orthocladiinae						8			8	c-g
f. Corynoneurina										
Corynoneura sp.						2				c-g
f.Ceratopogonidae										prd
Bezzia sp.			1			1				prd
f. Tipulidae										
Tipula sp.						1				prd
Pupae Chironomidae						1			4	
EPHEMEROPTERA										
Caenidae										
Caenis horaria						5				c-g

TAXON	STARČEVO			VIRPAZAR			PLAVNICA			FFG
TRICHOPTERA										
Polycentropodidae										
Poylocentropus sp.			3							prd
Decapoda										
Gammaridae										
Gammarus balcanicus			3							shr
Asellus aquaticus						1				shr
ODONATA										
Zygoptera (Coenagrionidae)										prd
Ischnura elegans						1				
COLEOPTERA										
Chrysomelidae										shr
Donacia sp.						1				
Total number of specimens	70	38	74	12	21	51	21	23	32	

Table 3 B. Structure of spring macrozoobenthic community. A-profundal part of lake, B-sub littoral part, C-littoral part. FFG-functional feeding group

8. References

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Fish Monitoring and Fisheries

Skadar/Shkodra

- Final Report -

August, 2014

Skadar/Shkodra

Final Report

Lead author:

PhD Danilo Mrdak, University of Montenegro, Faculty of Sciences and Mathematics, Depth of Biology

Co-authors:

NGO "HYDRA" team MSc Arian Palluqi, Prof. PhD Aleksandër Flloko,

PhD Eqerem Kapedani, Bardhyl Radovicka, Tom Miraku

PhD Dragana Milošević, University of Montenegro, Faculty of Sciences and Mathematics, Depth of Biology

Vukoica Despotović, University of Montenegro, Faculty of Sciences and Mathematics, Depth of Biology

ACRONYMS

AL – Albania

BPUE – Biomass per Unit Effort

CPUE – Catch per unit of effort

CSBL - Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Skadar/ Shkodra

FMO – Fishery management organization

Ha – Hectare (10.000 m^2)

IUU – Illegal, Unreported and Unregulated Fishery

MMG – Multi mesh size gill nets

MNE – Montenegro

SD – Standard deviation

SL – Skadar/Shkodra Lake

TL – Total length

TW – Total Weight

WFD – Water Framework Directive

Contents

1. Summary

MMG gillnets and EN 14757 standards implementation allows systematic approach in investigation of Skadar/Shkodra Lake fish fauna and fisheries on both side of the Lake. ON MNE side we detected 13 fish species while on AL side we detected 11 fish species in 2013 sampling campaign. On MNE side most abundant species were bleak (Alburnus scoranza), roach (Rutilus prespensis) and perch (Perca fluviatilis), respectively. On AL side most abundant were roach (Rutilus prespensis), bleak (Alburnus scoranza), perch (Perca fluviatilis), Pachychylon pictum and Pseudorasbora parva respectively.

In MNE part of the lake in deeper strata we detect less abundance but individuals were with larger body size (older age group cohorts) while for AL side of the lake we haven't detect such rule (for bleak and roach on AL side abundance was higher in deeper strata). MMG gill nets shows low capacity for netting of species with faster growing and with bigger body size (carp, chub, nase, mullet, prussian carp) while electro-fishing gives more detail insight in population structure of those species.

Proscribed minimal allowed length for bleak in MNE is completely wrong regarding that lowest allowed length for bleak is 16 cm. According to our data this length class is almost absent and biggest biomass of bleak is between 12 and 15 cm length.

Panels of MMG nets with mesh size range 19.5 -24 mm exclusively catch only bigger perch and bigger roach and shows not only selectivity for TL but also selectivity for only those two species.

Application of this monitoring scheme and methodology provide us a good basis regarding WFD needs. Diversification of sampling localities, gears in use and sampling period will allow more diverse sample in terms of fish species, especially those that are of importance for WFD.

Different composition of sampled species could be explained with differences in ecological parameters. Northwestern part of the lake is abundant in fresh water (one big inlet – Morača River, numerous middle and small size inlets and numerous external and sub-lacustrine springs and wells), characterized with divers Lake bottom substrate and whit vast flooding area along whole northern coast. Southeastern part of the Lake is mainly characterized with less impact of the fresh water, Lake bottom is mainly made of mud and stone, smaller flooding area and relatively closeness of marine habitats trough Buna/Bojana river as outlet. Fish species structure suggest that northwestern part of the lake is more oligotrophic than southeastern part which is caused by less impact of fresh water but also by more dense human settlements along lake coast (city of Shkodra as a major settlement).

Presence of trout in our samples suggests good ecological status of the Lake from WFD perspective. Major differences comparing with old fishery data from the past century are in rising of biomass and abundance of alien species (silver carp and perch) and domestic chub while on the other side we detected decreasing of biomass and abundance of nase, gray mullet and twaite-shad. In last several years in our research we haven't encounter mullet and nase in our samples (MNE side), and this research is something that encourages us to state that this year presence in the sampling of those two species point toward kind of Lake recovery. Future monitoring has to give precise answer whether this was some normal natural circle or it was trend that we detected. Our standing point is that final answer and more precise characterization toward fulfilling of WFD needs could be given only after at least 5 years of the same standardized monitoring (MMG nets, electro-fishing transects, larval seining, long-lines and monitoring of commercial catch).

2. Introduction

The aim of the CSBL project is to improve the implementation of legislation, regulations and management plans for the conservation and sustainable use of biodiversity at Lakes Prespa, Ohrid and Skadar/Shkodra. Component 2 of the CSBL plan of operation is targeting on fish monitoring and fisheries. Main tasks within this component are the implementation of joint monitoring for Lakes Prespa, Ohrid and Skadar/Shkodra, human, institutional and organizational capacity building, and fish and fisheries marketing, respectively. The data provide information needed for stock assessment and evaluation of stock dynamics of different species, particularly economic important fishes and species indicative for the ecological status of the lake. Also, data contribute to knowledge on aquatic biodiversity of lakes.

The agreed monitoring schemes is based on multi-mesh gillnetting according to the European Standard EN 147 57, as a central and common sampling element in all lakes. This kind of sampling is practiced for the first time on Skadar/Shkodra Lake. Additionally we performed three electro-fishing transect as well as monitoring of bleak harvesting in Raduš bay concessionary.

Regarding WFD, fish are one of the indicators for the ecological status of water bodies so the monitoring and status of their populations represent additional and important component for WF directive implementation related to water body classification.

Additionally, Skadar/Shkodra Lake is central water object for fresh water fishery in Montenegro and there is constant and strong market demand for Lake fish (carp, bleak, eel and chub mainly). Therefore there are numerous fishers on Lake that lives on finishing, fish processing and fish selling. Furthermore, Lake region is National Park in MNE while in AL it is region under special management and there are numerous fish restaurants along Lake coastal area that are representing one of the best known offer for tourist from MNE but also from AL. It is clear that fish and fish species are not only important because biodiversity and WFD issues, fish and their population in Lake are one of the most important economical resource for human population of Lake region.

3. Fish fauna and fisheries at Lake

The Skadar/Shkodra Lake drainage basin is located on the Montenegrin/Albanian border between 18º41’ and 19º47’ East and between 42º58’ and 40º10’ North. The lake is located in a karstic area in the outer part of the southeastern Dinaric Alps and is the largest of the Balkan lakes. It has a surface area which fluctuates seasonally from approximately 370 to 600 km^2, 60% of which belongs Montenegro and the remaining to Albania. The exact origin of the lake is unknown but it probably originated by solution and tectonic processes during the Pleistocene (Stanković, 1957). Lake is classified into the group of old Balkan lake, together with Prespa and Ohrid lakes. Those lakes in the same time represent model of old lakes of Europe, which are characterized with high level of endemism.

The main lake inlet is Morača river (average discharge of 202 m^3/s) while Bojana river is main Lake outlet (average discharge of 310 m^3/s). The rest (110 m^3/s) comes by precipitation, smaller tributaries and underwater karst springs called “vrulja” placed in Lake. The Skadar/Shkodra Lake is relatively shallow with mean depth of 5.01 m but there are locations where Lake depth go to 60 m in sublacustric springs called “oka” (Raduško oko). The lake bottom lay about 6 meters above sea-level and the lake coast line (with islands) is 207 km long. The Bojana river (Lake outlet) is a wide lowland river with length of 40 km which form huge estuary delta on Adriatic coast (near Ulcinj city). The Lake is placed in Mediterranean climate with average number of 130 “summer days” (days with temperature equal or higher then 25^o C). Mean value for precipitation is 2250 mm and there are no winter days (days with temperature equal or below 0^o C). The southern and south-western coasts (as well as narrow north-eastern part towards Rijeka Crnojevića) are steep mountains and hills slopes while northern and north-eastern coast is wide lowland area (swamps, flooded meadows, lowland forests...).

The Skadar/Shkodra Lake Region represents one of the most important centers of both, geodiversity and biodiversity for Western Balkan and SE Europe. The Lake region is a unique example of well-preserved freshwater ecosystems with specific geomorphologic, hydrological, soil and climatic features. It is also characterized by high biodiversity including numerous endemic and relict species of flora and fauna (Radović et al., 2008). The conditions for a diverse ichthyofauna in Lake are more than excellent. There are all kind of biotopes, lake have high level of bioproduction (thanks to shallow bottom almost all of Lake water-mass is in the photic zone) and high level of oxygen. Although the lake temperature is very variable (8-24^o C) there are locations where temperature of deeper part is more constant (Lake “oka”) what provides environment to steno-thermal organisms.

Ichthyic fauna of Skadar/Shkodra Lake is characterized by high number of its components. That includes number of freshwater and marine fish species, which periodically inhabit the lake. The structure of Lake Ichthyic fauna has been influenced by number of factors. First of all, there is geographic position and character of the lake, climate, physical and chemical characteristic of water and connection with rivers and Adriatic Sea. It has also been influenced by Ohrid-Drin-Skadar/Shkodra system, that connects lakes, river and sea ecosystem. River Bojana/Buna that drains from the lake is the main migratory road between lake and sea and allows migration of katadromous and anadromous fish species into the lake. Although there are no dams on this migratory rout, wire nets that occurs on Bojana/Buna River downstream from city of Shkodra significantly minimize migration possibility for both type of migration fish species. Presence of large number of sublacustric springs, so called “oka”, gives particular specificity to biology and ecology of ichthyic fauna components. That is particularly obvious in winter period, because “oka” are characterized by favorable thermal regime which makes them important winter habitats for some fish species. In the same time, those areas represent main points of harvesting of economy important fish species (dominantly Alburnus scoranza).

The number of fish species in Lake system increased in last 50 years because of uncontrolled stocking of alochthonous fish species which were originally from Black Sea drainage area (Hypophthalmichthys molitrix, Thymallus thymallus, Carassius gibelio, Perca fluviatilis...). But beside those introductions, the Skadar/Shkodra lake system (Skadar/Shkodra Lake and rivers in this water basin) is inhabited by several endemic species from families Salmonidae, Cyprinidae, Gobiidae and Cobitidae. The Lake is also spawning or growing zone for several anadromous and catadromous (migratory) fish species (e.g. Alosa falax, Anguilla anguilla, Acipenser sp.,) as well as for estuary fish species (Mugil cephalus, Dicentrarchus labrax, Platichthys flesus flesus...). Lake Skadar/Scadar/Shkodra, being at a low altitude, shallow, and warm, is an ideal cyprinid lake. But presence of small tributaries, big ones (Morača river on the first place) and sublacustric springs with relatively low and constant temperature allow salmonid fish fauna to live sympatric in the Lake (Salmo farioides and Salmo marmoratus). As a consequence, fish assemblage of Lake Skadar is characterized by a unique combination of cold and warm water species and an extraordinary species diversity.

Based on researches conducted so far, there are 50 species registered in Skadar/Shkodra lake. There are 37 native and 13 introduced fish species (Marić & Milošević, 2011). In the basin of Lake Skadar/Shkodra 7 fish species (18.3 %) are endemics. The endemics autochthonous fishes from Lake and its catchment area are: Salmo zetensis (Hadžišće, 1962); Barbatula zetensis (Šorić, 2000); Chondrostoma scodrensis (Elvira 1987); Gobio skadarensis (Karaman, 1936); Knipowitschia montenegrina (Kovačić & Šanda, 2007); Pomatoschistus montenegrensis (Miller & Šanda, 2008) and Rutilus albus (Marić, 2010). Natura 2000 list includes following species: Acipenser naccarii (Bonaparte, 1836), Acipenser sturio (Linnaeus 1758), Alosa falax (La Cepède, 1803), Rhodeus amarus (Bloch, 1782), Telestes montenigrinus (Vuković, 1963), Knipowitschia montenegrina (Kovačić & Šanda, 2007), Salmo marmoratus (Cuvier, 1829).

3.1. Fish fauna and stocks

By fish production and fish biomass Skadar/Shkodra Lake is far from other karst lakes on Balkan Peninsula. Based on statistical data of yearly catch, fish production in Lake is 80 kg/ha (8000 kg/km^2, data related to Montenegrin part of lake) (Drecun, 1983.). By this level of fish production, Lake is almost equal to some eutrophic lakes. Furthermore, Lake water is almost all time saturated with oxygen, there is no thermocline regarding its shallowness.

As well as in other bigger lakes, only restricted number of fish species dominates in normal fish production and therefore in total catch. Lake carp (Cyprinus carpio) and bleak (Alburnus scoranza) represent more than 70% of total catch. Beside those two species, Carassius gibelio, Alosa fallax, Anguilla anguilla, Chondrostoma nasus, Squalius platyceps, Scardinus knezevici, Rutilus prespensis and Mugil cephalus are also part of the catch.

In the Lake most of the fish is caught in fishing areas in northern, northwestern and southeastern parts as well as in Lake “oka” (Raduš, Krnjice, Ckla, Bobovište and Karuč in MNE). Major annual habitat shifts of most important species can be observed in spring and autumn. In spring, most of fish moved into littoral and flooded zone for spawn (cyprinid fish species). After spawning period, majority of adult fish moved from this flooded and littoral area in dipper pelagic region of lake as well as in deeper part along southern coast. With end of autumn majority of bleak began migration to their wintering habitats. During winter large concentration of fish are in lake “oka” (sublacutstric springs, cryptodepressions) where they spent winter.

Table 1. Fish species of Skadar/Shkodra Lake

Scientific name	English name	Native species	Introduced species (year of first introduction)	Species recorded in the past	Migratory type
Acipenser nacarii	Adriatic sturgeon			+	Temporary (anadromous species)
Acipenser sturio	Atlantic sturgeon			+	Temporary (anadromous species)
Alburnoides ohridanus	Ohrid spirlin	+			Al time present in the Lake
Alburnus scoranza	Bleak	+			Al time present in the Lake
Alosa fallax	Twait shad	+			Temporary (anadromous species)
Ameiurus nebulosus	Brown bullhead		+ (1978)	+	Al time present in the Lake
Anguilla anguilla	Eel	+			Temporally (catadromous species)
Atherina boyeri	Big-scale sand smelt	+			Temporary
Barbatula zetensis	Zeta stone loach	+			Al time present in the Lake
Barbus rebeli	Western Balkan barbel	+			Al time present in the Lake
Carassius gibelio	Prussian carp		+ (1973)		Al time present in the Lake
Chondrostoma scodrensis	Skadar nase			+	Al time present in the Lake
Chondrostoma ohridanus	Ohrid nase	+			Al time present in the Lake
Citharus linguatulus	Atlantic spotted flounder	+			Temporary
Cobitis ohridana	Ohrid spined loach	+			Al time present in the Lake
Ctenopharyngodon idella	Grass carp		+ (1975)	+	Al time present in the Lake
Cyprinus carpio	Carp	+			Al time present in the Lake
Dicentrarchus labrax	See bass	+			Temporary
Gambusia holbrooki	Mosquito fish		+ (1957)		Al time present in the Lake
Gasterosteus gymnurus	Western threespine stiklenek	+			Al time present in the Lake
Gobio skadarensis	Skadar gudeon	+			Al time present in the Lake
Hypophtalmichthys molitrix	Silver carp		+ (1973)	+	Al time present in the Lake
Hypophtalmichthys nobilis	Bighead carp		+ (1978)	+	Al time present in the Lake

Liza ramada	Thinlip mullet	+			Temporary
Megalobrama terminalis	Black Amur bream		+ (1973)	+	Al time present in the Lake
Mugil cephalus	Striped mullet	+			Temporary
Mylopharyngodon piceus	Black carp		+ (1983)	+	Al time present in the Lake
Oncorhynchus mykiss	Rainbow trout		+ (1951)		
Pachychilon pictum	Spoted roach	+			Al time present in the Lake
Pelasgus minutus	Ohrid minnow	+			Al time present in the Lake
Perca fluviatilis	Perch		+ (1978)		Al time present in the Lake
Phoxinus limaireul	Adriatic minnow	+			Al time present in the Lake
Pleuronectes flessus	Flounder	+			Temporary
Pomatoschistus montenegrensis	Skadar goby	+			Al time present in the Lake
Pseudorasbora parva	Stone moroko		+ (1977)		Al time present in the Lake
Rhodeus amarus	Bitterling	+			Al time present in the Lake
Rutilus albus	White roach	+			Al time present in the Lake
Rutilus prespensis	Roach	+			Al time present in the Lake
Salaria fluviatilis	Freshwater blenny	+			Al time present in the Lake
Salmo farioides	Adriatic trout	+			Al time present in the Lake (interdromous)
Salmo marmoratus	Marble trout	+			Al time present in the Lake (interdromous)
Salmothymus zetensis	Soft-muzzled trout	+			Temporary
Scardinius knezevici	Skadar rudd	+			Al time present in the Lake
Squalius platyceps	Shub	+			Al time present in the Lake
Telestes montenigrinus	Montenegro riffle dace	+			Al time present in the Lake
Tinca tinca	Tench		+ (1981)		Al time present in the Lake

3.2. Fisheries

3.2.1. Montengro

The majority of catch is taken with benthic nylon gill nets different in mesh size (so called bleak nets, carp nets, nase nets), with beach seines, with kalimera in concessionaries (Raduš and Karuč) and with lines (long line with lateral hooks). Large beach seines are almost exclusively used for fishing in lake "oka" and along lake shore during winter and spring period. Usually, heavy exploitation of fish occurs during shoaling period and in places where fish is concentrated. Maximum yield of bleak is during winter in lake "oka" and catch in that period (November – March) represents 76 % of total yearly Alburnus catch (Stein et al., 1981). Until 80's of past century when government proclaimed Skadar/Shkodra lake in National Park, maximum catch of Cyprinus occurred during March – May period (spawning time) in flooded and littoral lake areas where this species shoals for spawn.

The precise statistic of catch in Montenegrin part of Lake for last 40 years is completely absent but there are relatively confident data for catch in 1947th – 1976th period. In this period the total catch vary from 353 to 1311 tones (Figure 1.). The average yearly catch for 1953 – 1962 period was 1141 tons. This numbers should be increased for 10-15 % because there are no data related to other catch than those from registered fisherman who worked for public cooperative company. Of this 1141 tons of yearly catch, share of bleak was 570 tons, share of carp 230 tons, share of shad around 35 tons, share of eel, trout and mullet 25 tons, share of nase 30 tons while all other fish species were around 251 tons.

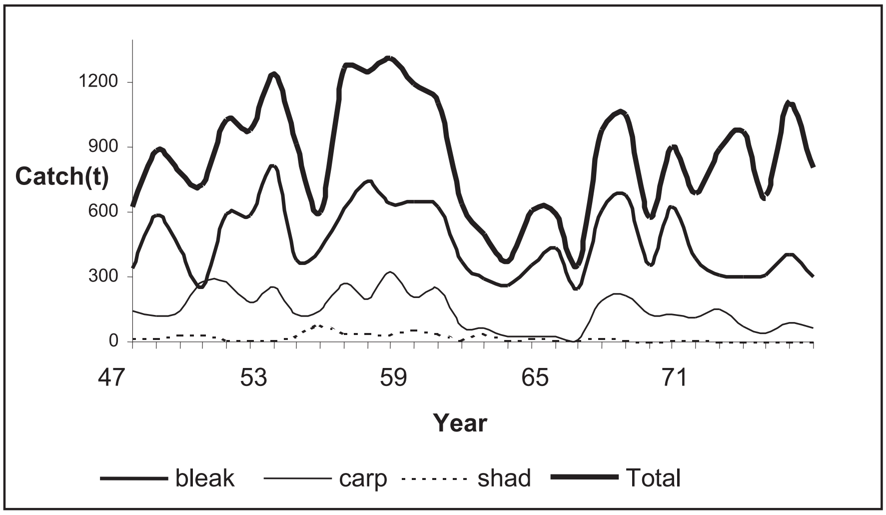


Figure 1. Catch statistic on Skadar/Shkodra Lake for 1947-1976 period (MNE side) (Stein et al., 1981)

During 70's of past century two new (introduced) fish species were noticed for Skadar/Shkodra lake, prussian carp (Carassius gibelio) (Vuković et al., 1975.) and perch (Perca fluviatilis) (Knežević, 1979.). The population of Prussian carp «explode» during 80's and 90's when this fish become statistically significant part of total catch. Unfortunately, the exact data of this species catch is missing but local fishermen are opinion that this fish, now, is third in total annual catch. Even more, some of those fishermen suppose that in 1985 – 1992 period Prussian carp was more abundant than common Lake carp. In present time, the population of Prussian carp is stabilized on significant lower level then during 90's and this fish take place of nase (Chondrostoma nasus) or even more, in Lake total annual catch.

The perch now represent main predatory species in Lake littoral area. After the introduction, this fish species found free ecological niche in Lake littoral area (before the introduction, only predatory species in Lake were salmonids and eel). The perch population grows dramatically and nowadays it is one of most abundant fish species in Lake littoral parts. In last two decade of 20th century the new species was assorted in "scrap" fish without of strong market demands. That was because perch wasn't present in Lake and people rather bought domestic species (carp, bleak, chub, rudd, roach...). In last 10 years people become familiar with perch and start to consume this species, what produce market demands for this fish.

In last 20 years there was no well-organized research on fishery issue and no continues monitoring of fishery in Lake. In this period there were only individual research mainly related to fish taxonomy. During the 90's fishing activities on Lake was without any restriction (there was positive legislation related to fishing activities, mesh size, ban of fishing activities during spawn period, forbidden fishing tools...but there was almost no implementation of this legislative). In last 10 years the management of National Park strictly put in use all fishery legislation and rules. They again reestablish fishing license (for recreational and professional anglers) and start with controlling and arresting of poachers. But again there is no data related to catch.

3.2.1.1 *Management and legislation*

In Montenegro Skadar/Shkodra Lake has been a National Park since 1983. In 1995 National Park "Skadar/Shkodra Lake" was recorded in the Ramsar list (wetland area of international significance).

According to the Article 25 of the Law on National Parks (Official Gazette 47/91) and the Statute of the Public Enterprise for National Parks of Montenegro, in 2005, the Board of the company passed the Decision on requirements for commercial fishing on Lake, which defines the parts of the Lake where commercial fishing shall be carried out.

It has been stipulated in the respective Decree that commercial fishing shall be carried out in the all surface of Lake, except in designated places, listed as follows:

- **In the locations called Crni zar, Pančeva oka, Omerova gorica, Manastirski vrbiš and Grmozur,**
- **On the location called Tanki Rt at 500 m on either side of the bridge,**
- **Downstream the river to the places called Crnojevića Gradina and Lisinj.**

Commercial fishing can only be carried out with registered fishing boat, motor power up to 10 HP and visibly marked fishing equipment. Carp and other fish species may be catch with a maximum of 20 net whose stretched mesh size is not below the eyelets of 60 mm, bleak with a maximum of 10 bleak net whose diameter is not below the 15.5 mm eyelets, eel and carp of up to 10 fyke-nets whose diameter is not below 25mm eyelets, and with long-lines with a maximum of 500 hooks.

Relevant laws and bylaws:

- **Law on National Parks (Official Gazette of the RM, No.47/91, 27/94)**
- **Environmental Law (Official Gazette of the RM, No. 12/96, 55/00)**
- **Environmental Impact Assessment Decree (Official Gazette of the RM, No.14/97)**
- **Law on Nature Protection (Official Gazette of the RM, No. 36/77,2/89)**
- **Decree on Protection of Rare, Scarced, Endemic and Endangered Plant and Animal Species (Official Gazette of the RM, No.36/82)**
- **Law on Freshwater Fishery, (Official Gazette of the RM, No. 39/76, 51/76, 34/88, 29/89, 39/89, 48/91, 4/92, 17/92, 27/94)**
- **Law on Waters (Official Gazette of the RM, No. 16/95)**
- **Decree to Prohibit the Use of Vessels with Engine Power more than 4.5 HP by physical Persons On Skadar/Shkodra Lake (Official Gazette of the RM, No.9/86)**
- **Decision on the Level and the Method of Payment of Charges for the Use of National Parks Assets, for Conducting Economic Activities and Providing Services (Official Gazette of the RM, No.31/02)**
- **Law on the Protection of Cultural Monuments (Official Gazette of the RM, No.47/91)**
- **Law on Local Self-Government (Official Gazette of the RM, No.75/05)**
- **Law on Inspection (Official Gazette RCG, No.50/92).**

3.2.2. Albania

The fishery sector in Albanian part of lake Skadar/Shkodra is quite important. According to the last census done in the Albanian side there are 210 boats used by 410 fishers that operate in the Albanian side, with the main fishers are located in Shiroka and Zogaj villages and near Koplik city. The main species caught for commercial purposes are bleak and carp, although in specific seasons, eel and mullet become important due to the connection of the lake with the sea. The fishers are organized in a FMO which is increasing its capacities and is trying to collect most of the fishery product coming from fishers and recently is using some facilities for processing fish (mainly drying and smoking).

The majority of catch is taken with benthic nylon gill nets different in mesh size (so called bleak nets, carp nets, nase nets), with beach seines, and with lines (long line with lateral hooks). Large beach seines are used in the winter period for the bleak fishery in two localities (Shiroka and Zogaj). The winter period is the season where the majority of the catch is bleak while in other seasons other species become more relevant. For the last 25 years no reliable data exist on the catch in Lake since no real collection of data were available. The most recent is the one done in 2011, where all fishers present in the Albanian side (registered in the FMO) fulfilled logbooks for their activity. From the calculations, approximately 500 t are caught each year in the Albanian side (Figure 2) while the main species are bleak, carp and roach (for roach fishers has clustered several species in the logbook). Another big issue is the IUU fishery, especially electric fishery which may account for a large part of the catch in Albania. Furthermore, data from the fish weir, which are especially important from the migratory species are not accurate and not reliable.

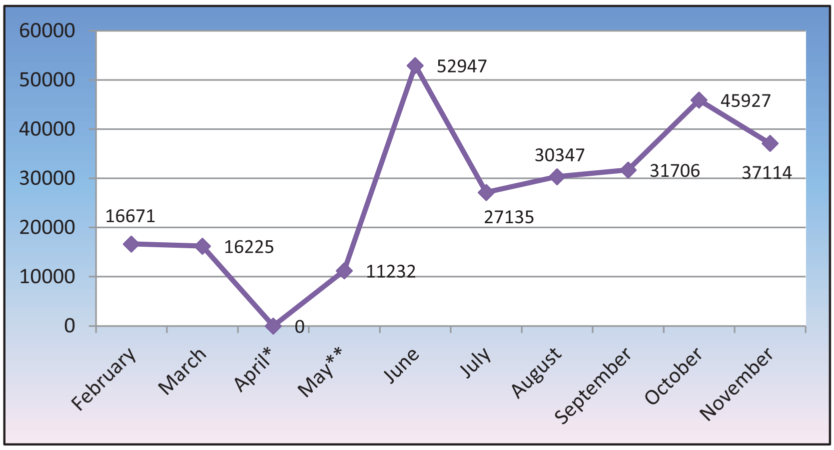


Figure 2. Catch statistic on Al side based on 2011 census (registered fisherman logbooks) * and ** - April and half of May are closed season for commercial fishery

The catch was composed mainly of 7 species (although fishermen classify as roach not only one species). The catch composition varies greatly depending on the season (Figure 3)

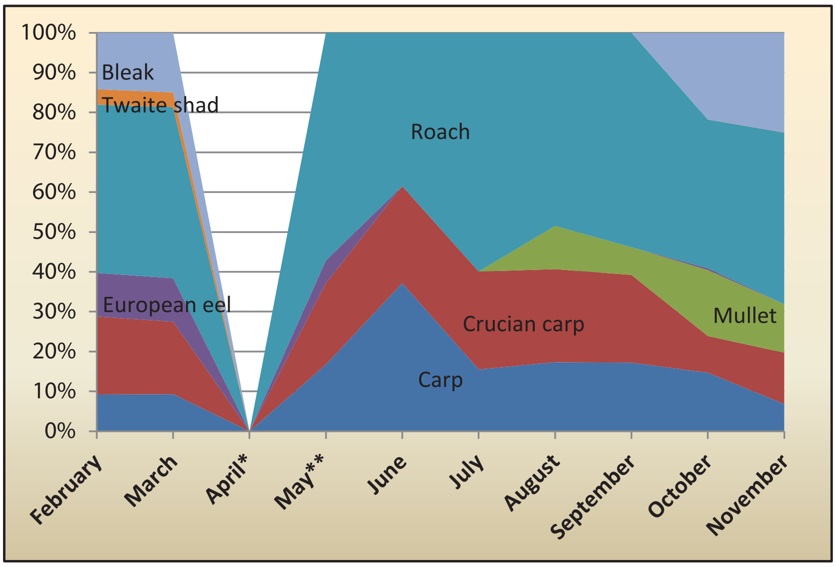


Figure 3. Species composition in commercial catch in 2011th (* and ** - April and half of May are closed season for commercial fishery)

Table 2. Composition of species in total commercial catch in 2011th (in kg)

	Feb.	Mar.	Apr.*	May**	June	July	Aug.	Sept.	Oct.	Nov.
Carp	1536	1510		1880	19673	4208	5252	5454	6743	2531
Crucian carp	3260	2947		2286	12891	6664	7090	6983	4228	4779
Mullet	0	0		0	0	0	3296	2199	7599	4512
European eel	1825	1772		648	0	0	0	0	215	0
Roach	7039	6965		6418	20383	16263	14709	17070	17134	15993
Twaite shad	649	600		0	0	0	0	0	0	0
Bleak	2362	2431		0	0	0	0	0	10008	9299
Total	**16671**	**16225**	**0**	**11232**	**52947**	**27135**	**30347**	**31706**	**45927**	**37114**

3.2.2.1 Management and legislation

In Albania the actual protection status of Skadar/Shkodra Lake is "Managed Natural Reserve" (IUCN Category IV), declared by the Albanian Government with the Decree of Council of Ministers No. 684, of 02.11.2005.

The protected area in Albanian side of the lake includes three categories of protection:

a) The core zone composed by: the lake shore from the western extreme of Zogaj village to the border between Republic of Albania and Montenegro, the slope of Taraboshi Mountain from altitude 494 m in the south to 200 m within the lake waters, in the segment Zogaj-Albanian-Montenegrin border in the north. The second level of protection is applied in this area.

b) Habitat Management Area composed by: the whole lake water surface, except the one included in the area 1/a mentioned above; the Albanian western shore from Bojana/Buna bridge in the east, to Zogaj village in the west (bordering zone 1/a of this point) including all the latitude of this segment up to the altitude of 300 m in Taraboshi mountain slope in the south. The third level of protection is applied in this area.

c) Traditional Development Area composed by: the whole eastern surface of the lake bordering on the west with the area 2/a mentioned above of this point up to Shkodër-Hani i Hotit motorway in the east and Shkodra city in the south-eastern end. The fourth level of protection is applied in this area.

The Albanian part of the lake area has been also declared as a Ramsar site (Ramsar Convention "On internationally important wetlands, especially as waterfowl habitats") by the Ramsar Secretariat and the Decree of Council of Ministers No. 683, of 02. 11. 2005 of the Albanian Government. There are some recent joint efforts being taken to declare lake Shkodra/Skadar /Shkodra as UNESCO Biosphere Reserve.

The legislation framework in the fishery sector, in an overall overview is complete and contemporaneous. The legislation deals not only with fishery issues but also with other related issues such as: biodiversity, socio-economic aspects etc.

The legislation framework includes all levels of legal and normative acts, such as laws, by-laws, Regulations, Decrees of The Council of Ministers, Normative Acts etc.

There are several legislative acts regulating the fishing activity, including:

The new **Law No. 64/2012** set the basis for the good management of the fishery sector explains many of the terms and concepts related to the fishery sector

It should be stressed that the main intention of the law are:

- **to ensure a rational and accountable exploitation of aquatic biological resources and development of aquaculture;**
- **provide protective conservation measures in order to ensure the protection of biological water resources, and**
- **support the sustainable development of fishery and aquaculture sectors, as well as create better socio-economic conditions for producers.**

Based on the above regulations, in the Albanian side, fishery sector in Skadar/Shkodra Lake is regulated as follow:

Each boat is permitted to have 2000 meters of entangling nets (of which 1000 meters of trammel nets and 1000 meters of gillnets). For each trammel net, 500 meters have 80 mm mesh size and 500 meters with 35-40 mm of mesh size. Meanwhile the same length (500 meters each) and the same mesh sizes respectively are used for gillnets.

The number of hooks for each boat has not been established yet. There are two seiners with 28 mm of mesh size for the bleak fishery in winter situated one in Shirokë and one in Zogaj. Three other seiners are for carp and crucian carp used in Shirokë, Zogaj and Kamicë. Shirokë and Zogaj has also two sorting boxes for the selection of bleak and carp.

Relevant laws:

- **Law "On the Land" (1991)**
- **Law "On Forests and Forestry Police" (1992)**
- **Law "On Protection of Wild Fauna and Hunting" (1994)**
- **Law "On Fishing and Aquatic Life" (1995)**
- **Law "On the Regulatory Framework of the Water Supply Sector and of Disposal and Treatment of Waste Water (1996)**
- **Law "On Protected Areas" (2002)**
- **Law "On Protection of Marine Environment from Pollution and Damage" (2002)**
- **Law "On Protection of Trans-border Lakes" (2003)**
- **Law "On Environment Protection" (2011)**
- **Law "On Environmental Impact Assessment" (2011)**
- **Law "On Integrated Management of Water Resources" (2011)**

3.3. Comparative review of fishing/fishery rules in MNE and ALB

In following tables we give a comparative review of main fishing/fishery rules on both side of the Skadar/Shkodra Lake:

Table 3. Fishing ban season by species and by countries

SPECIES	Montenegrin part		Albanian part	
	from	to	from	to
Carp	15th March	1st June	15th April	15th May
Chub	15th March	1st June	15th April	15th May
Nase	15th March	1st June	15th April	15th June
Roach	15th March	1st June	15th April	15th June
Bleak	15th March	31st October	1st April	31st July

In MNE additionally is proscribed fishing ban season for trout which is from 1st October to 1st April.

Table 4. Minimum allowed dimensions for fishing of some commercial species

Common name	Scientific name	Montenegro	Albania
Carp	*Cyprinus carpio*	40 cm	30 cm
Chub	*Squalius spp*	Not specified	15 cm
Prussian carp	*Carassius gibelio*	Not specified	15 cm
Nase	*Chondrostoma spp*	Not specified	15 cm
Roach	*Rutilus prespensis*	Not specified	12 cm
Bleak	*Alburnus scoranza*	16 cm	10 cm
Perch	*Perca fluviatilis*	Not specified	15 cm
Brown trout	*Salmo farioides*	25 cm	Not applicable
Mullet	*Mugil spp.*	25 cm	Not specified
Marble trout	*Salmo marmoratus*	50 cm	Not applicable
Lake trout	*Salmo farioides*	30 cm	Not applicable

4. Trans-boundary sampling scheme performed

4.1. Methodology of sampling

Table 5. Sampling scheme and gears suggested for sampling on Skadar/Shkodra Lake

	Montenegro												Albania											
METHOD	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
MMG (EU STANDARD, EN 14757)									■	■	■					■	■	■					■	
Fyke Net									■	■	■					■	■	■					■	
Comm. Beach Seine	■	■											■											■
Electro-fishing transects		■							■															
Monitoring of commertial harvesting (Rads bay)	■	■										■												
Experimentl Beach seine(larva)				■	■	■			■							■	■	■			■			
Catch data	■	■	■			■	■	■	■	■	■	■	■	■	■			■	■	■	■	■	■	■

Sampling scheme was agreed on second fish/fishery group meeting held in city of Shkodra. From suggested methods we haven't accomplished following: experimental larval beach seining (both country), commercial beach seining (both country), catch data (both country). The reason for not accomplishing of beach seining is that we were depending on fishermen who are obliged to announce commercial beach seining (position and data) to fishery management authorities and according to authorities in both country they haven't announced any in 2013/2014 winter period. Since we haven't our own beach seining net we couldn't accomplished this task. As for experimental larval beach seining we had problems with delivery of gear which was delivered in late November when there was no fish larva (spawn occurs in spring and up to November young fish grows in to recruitments stage). Monitoring of commercial catch hasn't been done in MNE since fishermen in MNE are not obliged to fulfill logbooks of catch. Although in AL fishermen are obliged to fulfill the logbooks, still there is no system in place, so we couldn't collect data. In 2011th in Albania data collection was supported by other project and three persons were paid for data collecting and that is how we have data for this year. Fyke - nets were posed in both country but regarding need of avoiding periods with fish movement and aggregation we were forced to conduct our research in September-November period. Basic purpose of fyke-net posing was sampling of eel but in all our attempts we haven't any in our fyke-net traps. Therefore we didn't show those data since we failed in eel sampling (wrong sampling season for eel).

In order to apply EU standard researching nets (multy-mesh size gill nets – **MMG**) we follow EN 14757 standard in terms of time, number and stratification of sampling. We were faced with problem regarding this standard since it doesn't have any recommendation for huge lakes such is Skadar/Shkodra Lake so we were forced to do some approximations. We chose three parts of the Lake (one big part of the Lake on MNE and two smaller parts on AL side of border) end performed sampling scheme according to EN 14757 standard. In order to follow the basic principle of representative sampling we avoided periods of fish grouping (spawn and wintering shoaling). For second sampling methodology (electro-fishing) we also avoid periods of fish shoaling. Considering relatively shallow MNE part and almost no strata that is below 6 m of depth we sample with 24 MMG gillnets. In Albanian side we choose two sectors with three strata (0-3, 3-6 and >6m of depth) and therefore we sample with 36 nets on each sector (Figure 4 and 5).

According to recommendation of project scientific supervisor for fishery we choose one big area of Lake in MNE (2 km x 2,5 km, Vranjina/Virpazar/Grmožur sub-basin) and two smaller areas in Albania (Shiroka and Koplik sub-basins). In MNE part we had two different depth strata, smaller shallow stratum 0-3m depth (about 30 % of sub-basin area) and deeper 3-6m stratum (about 60 % of sub-basin area) while in Shiroka and Koplik areas (AL) there were three strata: 0-3m stratum, 3-6m stratum and more than

6m m stratum. On MNE side such Vranjina/Virpazar/Grmožur sub-basin is composed of all fish habitats in terms of submerged, emerged and floating vegetation as well as in terms of different type of bottom (gravel, rocky and muddy substratum). As for Shiroka sub-basin, dominant was rocky substratum while for Koplik it was dominant muddy substratum. In both sub-basins, like for the whole lake, muddy bottom was dominant in deeper parts.

Figure 4. Sampling scheme for MNE part of the lake for MMG nets and Electro-fishing (yellow dots and numbers – 0-3m stratum MMG nets, red dots and numbers – 3-6m stratum MMG nets, green lines -transects, T1, T2 and T3)

Figure 5. Sampling scheme for AL part of the lake (MMG nets, 1 Shiroka and 2 Koplik sector)

4.1.1. Multimesh gillnetting (MMG nets)

We use standard benthic MMG nets that are 30 m long, 1.5m high and made of 12 different panels with different mesh size diameter (so called "knot-knot" measure). Mesh size ranged between 5 and 55 mm in geometric row.

Multimesh gillnets in MNE part were posed in period of 8.10.- 9.11.2013 in 6 series, 4 nets per each serial. Sampling scheme was defined in order to cover both strata in sampling area, 0-3m strata (yellow dots on Figure 4) and 3-6m strata (red dots on Figure 4.). Nets were posed randomly. In total there has been 24 benthic gillnets posed randomly but keeping in mind that shallower zone represent 30 % while deeper zone is 60 % of total area (8 nets for 0-3 strata and 16 nets for 3-6m strata, Figure 4). Nets were posed in the evening and pulled out in the morning so the each net was fishing 12 hours over night.

Multimesh gillnets in AL part were set in period of 27.10-25.11.2013 in two distinctive areas, Koplik and in Shiroka (Figure 5). The two areas represent separate regions of the lake (south part of the lake and eastern part of the lake) as shown in Figure 5

The 72 benthic nets were randomly set in the different depths. The directions of the nets were (i) parallel with the lakeshore, (ii) perpendicular with the lakeshore and (iii) at 45 degrees with the lakeshore. In Koplik sector there were 15 nets in the 0-3 m stratum, 12 nets in the 3-6 m stratum and 9 nets in the more than 6 m stratum. In Shiroka sector there were 13 nets set at the 0-3 m stratum, 11 nets at the 3-6 m stratum and 12 nets at the more than 6 m stratum. Nets were posed in the evening and pulled out in the morning so the each net was fishing 12 hours over night.

All netted fish were determined to species level, each fish was measured in terms of Total Length (TL) and Total Weight (TW) for each panel of each MMG net. Such data were noted for each individual net and later written in previously prepared Excel file for further data processing. Such data were record for each net and panel as well.

4.1.2. Electrofishing

Electrofishing transect were performed in MNE part of the lake in 0-3m strata (green lines, T1, T2 and T3, Figure 2). We use standard electrofishing gear for boat, one person perform electrofishing while other two collect fish with two hand-nets. Transect were 500m long while electrofishing gear cover 2m on both size, so the transect was 500m long and 4m wide (in total 2000 m^2). In order to cover different habitats we performed electrofishing transects in three different localities of the investigated are. First one was nearby Morača river mouth (sandy-muddy bottom, strong impact of fresh riverine water) second one was on southern Lake coast (rocky to muddy bottom with moderate plant coverage) while third one was in standard lake bay (muddy bottom with intensive plant coverage) (Figure 2.). Each fish was determined to species level measured (TL and TW) and data were written in previously prepared Excel sheet file for further data processing.

4.1.3. Sampling of commercial bleak harvesting (Raduš bay)

In MNE in Raduš bay, during wintering period (December, January and February) it occurs industrial harvesting of bleak which is performing by the concessioners. For this purpose they use so called "kalimera" net which is essentially big net (30 x 30 m) posed horizontally in water and which harvest bleak during extraction in vertical column above it.

We take subsample from the total catch, approximately one bucket of fish from the center of harvested individuals (8 kg of fish). Each fish was determined to species level measured (TL and TW) and data were written in previously prepared Excel sheet file for further data processing.

4.2. Data analysis and management

4.2.1. Multimesh gillnetting (MMG nets)

CPUE values (Catch per unit of effort) was calculated for each net and each netted species as a relive measure of species abundance. CPUE was calculated as follows: total catch in grams of individuals of each species in MMG net (all panels) divided with net surface (1,5m X 30m = 45m^2, CPUE grams) and total catch in number of individuals of each species in MMG net (all panels) divided with net surface (1,5m X 30m = 45m^2, CPUE individuals). Since all nets were fishing approximately 12 hours we haven't include this in CPUE calculation so the CPUE values are in [gr/m^2] and in [No of ind./ m^2] not in [gr/m^2/hours of fishing] and not in [No of ind./m^2/hours of fishing] .

The average CPUE value for each species in each strata (0-3m, 3-6m and >6m stratum) was calculated as follows:

- **Sum of each net CPUE value of species *N* / number of nets in strata**

4.2.2. Electrofishing

Since the Electro-fishing is more selective than MMG nets it is usually used for fish inventory (fish community composition) research but here on Skadar/Shkodra Lake our electrofishing samples shows just opposite selectiveness comparing to MMG nets. Namely, MMG nets samples shows almost no bigger body size fish species while electrofishing was more effective on such fish species (with electrofishing we more commonly got bigger fish size regarding their bigger volume and more effective impact of electro-magnetic field on it). That is why we decide to calculate CPUE for electrofishing transects end we think that this value is also comparable if we repeat it every year just like CPUE form MMG nets.

CPUE values for electrofishing transect were calculated as a Total weight of species X [gr] divided with 2000m^2 (transect surface, 4m x 500m). For big body size species (carp, chub, nase and Prussian carp) we construct Length/Weight graph together with growing curve (Annex 1). For smaller sized fish species and those that were highly frequent in MMG net samples we haven't construct Length/Weight graph and growing curve out from electrofishing transect data sets.

4.2.3. Sampling of commercial bleak harvesting (Raduš bay)

As we take a subsample from "kalimera" net and considering that this is highly selective fishing gear used for commercial harvesting during winter shoaling we use data from those subsamples to analyze the structure of catch (in terms of weight and in terms of diversity) as well as to analyze the selectivity of "kalimera" net. Therefore we construct length frequency of bleak from subsamples in order to analyze length cohort proportions in commercial "kalimera" catch.

4.3. Results

4.3.1. Country: Montenegro

4.3.1.1 Multimesh gillnetting

According to standard EN 14757 we choose part of the Skadar/Shkodra Lake with surface of 5 km^2 and we pose 24 gillnets as it is suggested for lake with average depth not more than 6 m. Nets were posed randomly in two depth strata: 0-3m strata (8 MMG gillnets) and 3-6m strata (16 MMG gillnets) (Figure 4.).

As we mentioned in methodology part, data were processed and analyzed in separate clusters (two strata) and the following results are presented as follows.

4.3.1.1.1. Abundance

Table 6. Number of individuals per each net per each sampled species for both strata

	Alburnus scoranza	*Rutilus prespensis*	*Perca fluviatilis*	*Scardinius knezevici*	*Pseudorasbora parva*	*Carassius gibelio*	*Pachychilon pictum*	*Rutilus albus*	*Squalius platyceps*	*Mugil cephalus*	*Chondrostoma nasus*	*Cyprinus carpio*	*Rhodeus amarus*	**SUM per net**
0-3m stratum														
Net 1	180	69	60	4	1	1	1	2	1					**319**
Net 2	18	82	15	10	6			4	1	1	1			**138**
Net 3	93	72	118	1	1	6	1	1				2		**295**
Net 4	173	28	33	1		2								**237**
Net 5	96	43	22		3			1						**165**
Net 6	72	34	15		1									**122**
Net 7	70	31	16		2		1	1			1		1	**123**
Net 8	60	63	35	1								1		**160**
SUM per species	**762**	**422**	**314**	**17**	**14**	**9**	**3**	**9**	**2**	**1**	**2**	**3**	**1**	**1559**
3-6m stratum														
Net 9	5	21	13			1								40
Net 10	6	15	9			2								32
Net 11	21	28	12											61
Net 12	31	41	10						1					83
Net 13	10	4	11		2			1						28
Net 14	7	13	25		2									47
Net 15	10	12	23											45
Net 16	7	18	13											38
Net 17	22	60	6					4						92
Net 18	36	22	13			1						1		73
Net 19	90	66	16		4	2			1					179
Net 20	129	52	21	2	3				1					208
Net 21	8	24	7			1								40
Net 22	10	4	5									1		20
Net 23	36	26	8		3									73
Net 24	19	8	5											32
SUM per species	**447**	**414**	**197**	**2**	**14**	**7**		**5**	**3**			**2**		**1091**

In 0-3m of depth stratum we detected 13 fish species. Alburnus scoranza was fare more dominant with 49 % of all sampled individuals. Together with Rutilus prespensis and Perca fluviatilis takes 96% of all sampled individuals (Figure 6.).

In 3-6m stratum we detected 9 fish species while the dominance of bleak (Alburnus scoranza) was not as obvious as for 0-3m stratum. Namely bleak takes 41 % while roach (Rutilus prespensis) takes 38 % of total sampled individuals. Again those three species (bleak, perch and roach) are dominant representing 97 % of all individuals in sample (Figure 7).

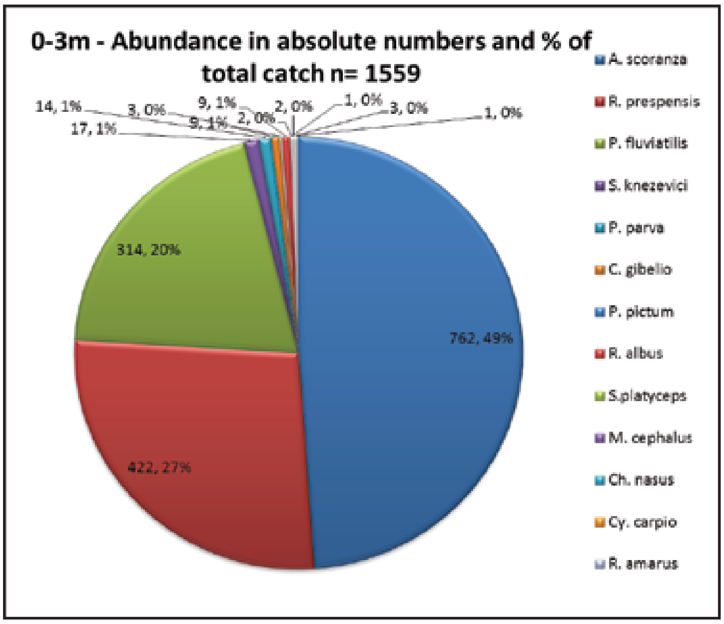


Figure 6. Abundance of recorded species in absolute number and percentage of total individuals number (0-3m strata) (absolute number, percentage).

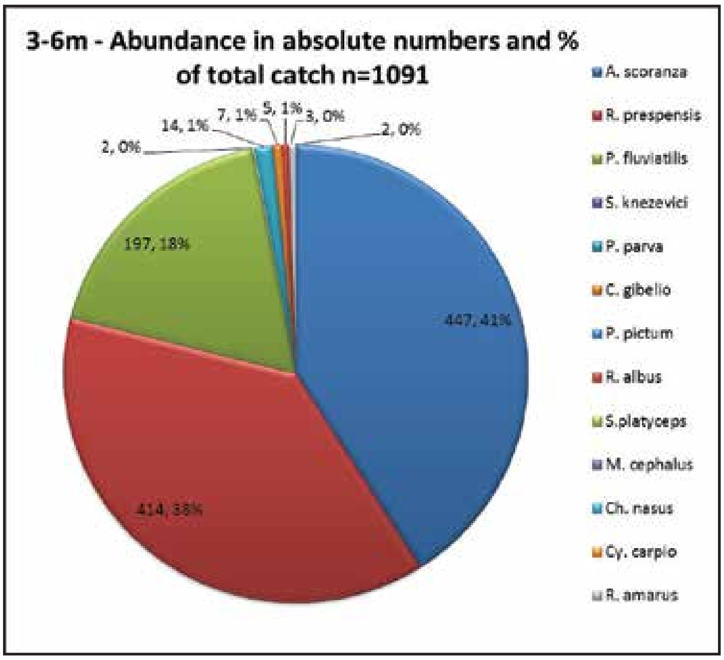


Figure 7. Abundance of recorded species in in absolute number and percentage of total individuals number (3-6m strata) (absolute number, percentage).

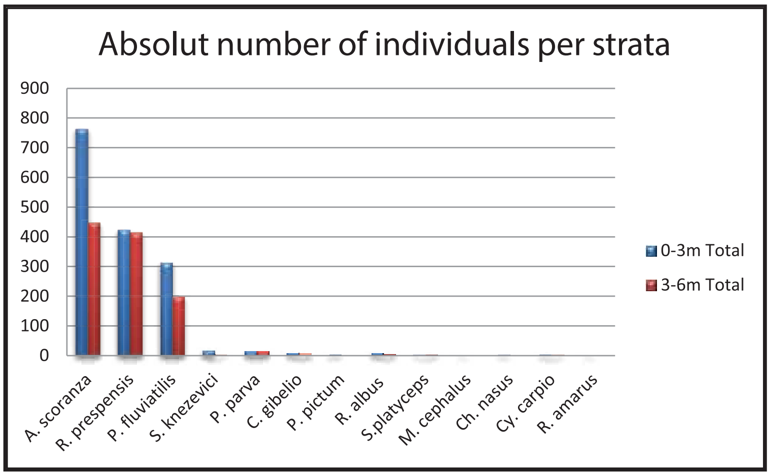


Figure 8. Absolute number of individuals for both strata

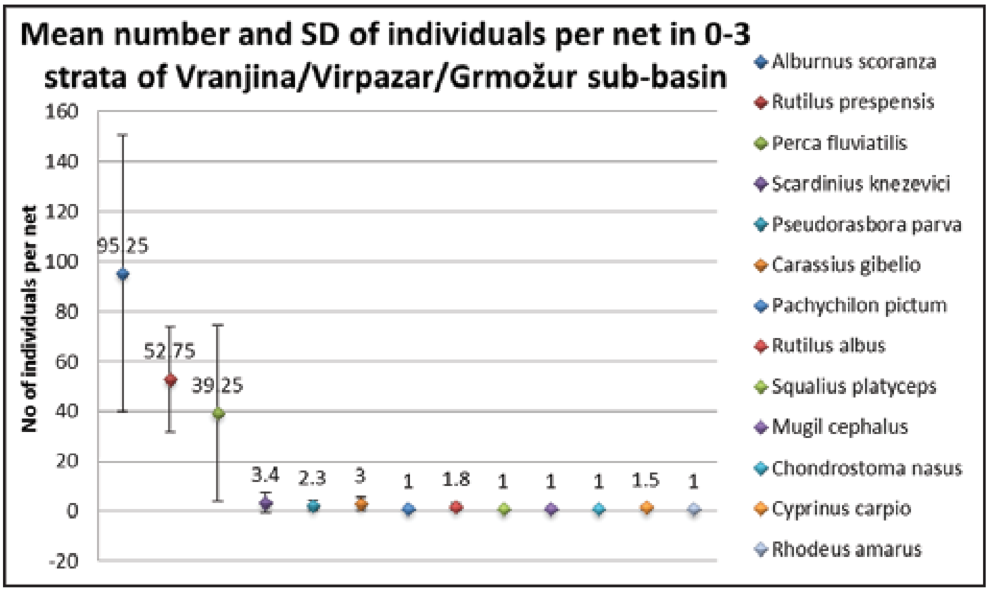


Figure 9. Mean number of individuals and standard deviation in one MMG net for 0-3m stratum

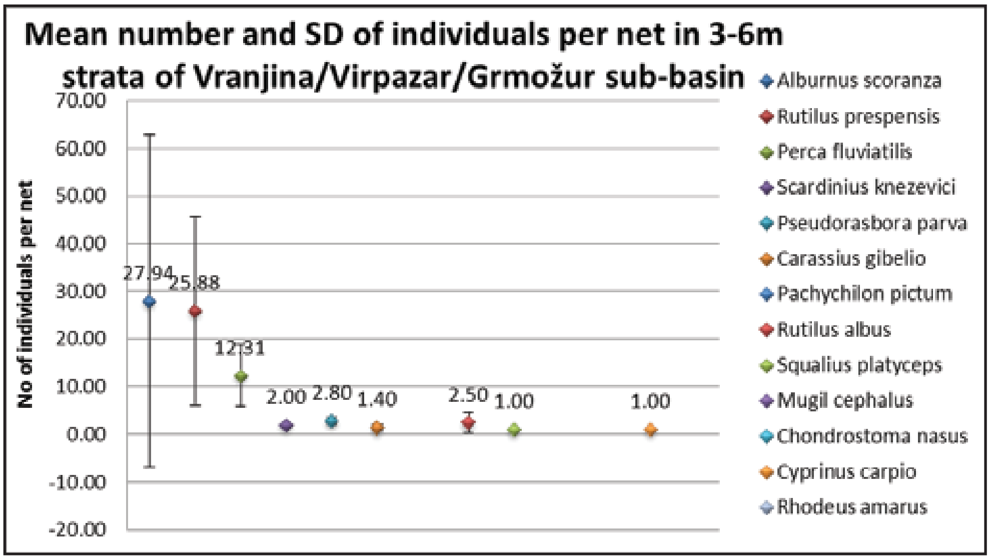


Figure 10. Mean number of individuals and standard deviation in one MMG net for 3-6m stratum

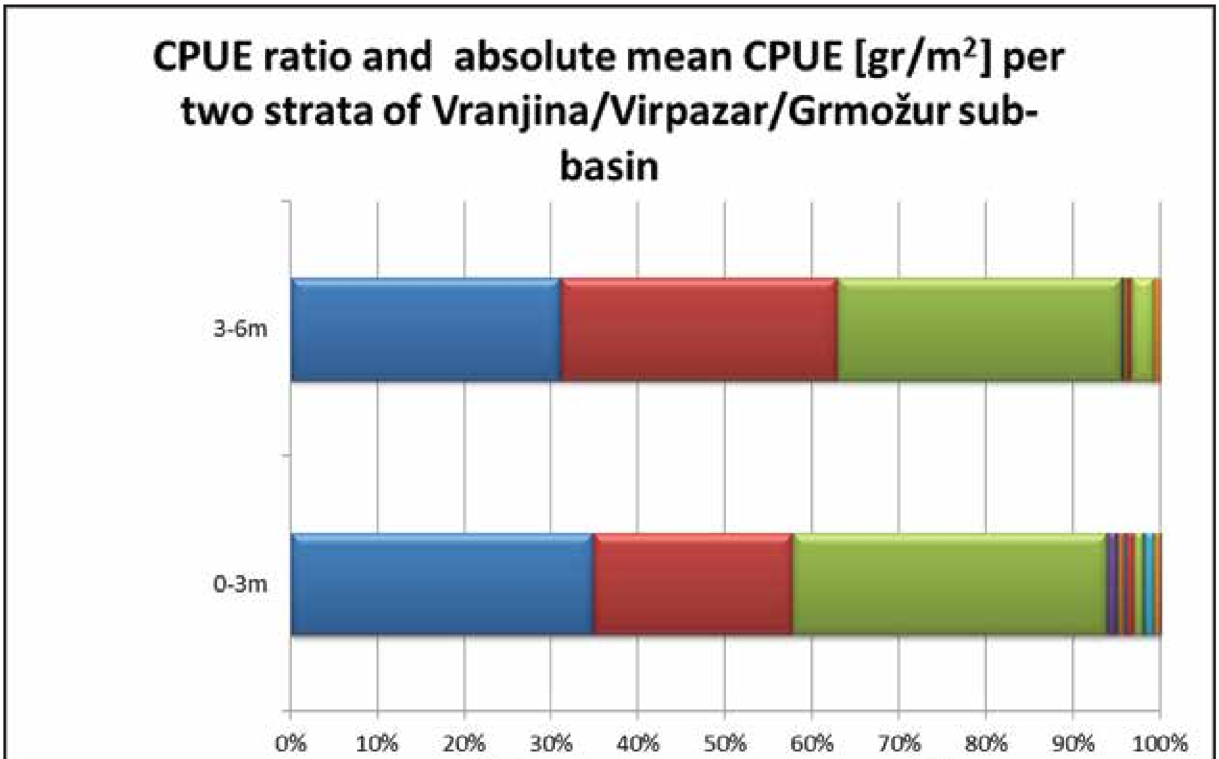
Table 7. Average number of individuals and SD per one MMG net for both strata

	Alburnus scoranza	*Rutilus prespensis*	*Perca fluviatilis*	*Scardinius knezevici*	*Pseudorasbora parva*	*Carassius gibelio*	*Pachychilon pictum*	*Rutilus albus*	*Squalius platyceps*	*Mugil cephalus*	*Chondrostoma nasus*	*Cyprinus carpio*	*Rhodeus amarus*
	0-3m stratum												
Average No of individuals/net	95.25	52.75	39.25	3.4	2.3	3	1	1.8	1	1	1	1.5	1
SD	55.58	21.14	35.26	3.91	1.97	2.65	0	1.30	0	0	0	0.71	0
	3-6m stratum												
Average No of individuals/net	27.94	25.88	12.31	2.00	2.80	1.40	0	2.50	1.00	0	0	1.00	0
SD	34.90	19.86	6.42	0.00	0.84	0.58	0	2.12	0.00	0	0	0.00	0

Although we had two times more nets posed in deeper stratum absolute number of individuals of three dominant species was higher in shallow stratum suggesting denser abundance (in terms of number of individuals per standard surface) (Figure 8.). This is more obvious if we analyze mean number of individuals per one MMG net (bleak, perch and roach) where we had twice more individuals in 0-3 than in 3-6m stratum (Figures 9., 10. and Table 7.).

4.3.1.1.2. CPUE

Following figure shows CPUE values ratio for all sampled species per both strata (Figure 7.). Calculated CPUE values shows that catch of bleak and perch (in terms of gr/m^2 of total MMG net) has bigger proportion in 0-3m stratum while roach has opposite tendency (bigger proportion is in sample from 3-6m stratum) (Figure 10). Bigger size species such are Squalius platyceps (chub) and Cyprinus carpio (carp) has bigger proportion of total catch in deeper stratum (3-6m).



	0-3m	3-6m
Alburnus scoranza	37.998	15.146
Rutilus prespensis	25.128	15.600
Perca fluviatilis	39.488	15.995
Scardinius knezevici	1.219	0.071
Pseudorasbora parva	0.141	0.091
Carassius gibelio	0.704	0.213
Pachychilon pictum	0.373	0.000
Rutilus albus	0.934	0.203
Squalius platyceps	1.141	1.264
Mugil cephalus	0.169	0.000
Chondrostoma nasus	1.204	0.000
Cyprinus carpio	0.762	0.346
Rhodeus amarus	0.004	0.000

Figure 11. CPUE ratio and absolute average CPUE [gr/m^2] per two strata of Vranjina/Virpazar/Grmožur sub-basin

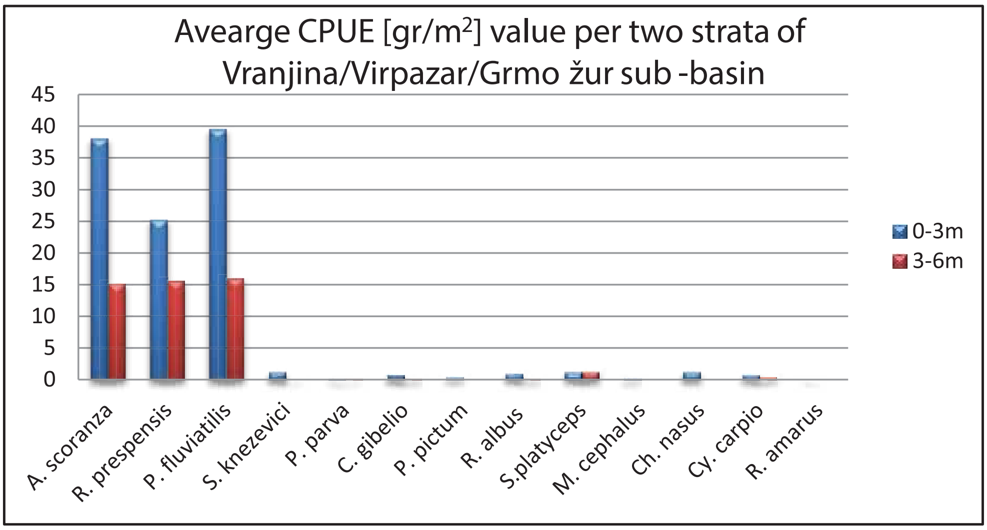


Figure 12. Comparative review of calculated average CPUE values (gr/m² of MMG net) for all sampled species in both strata

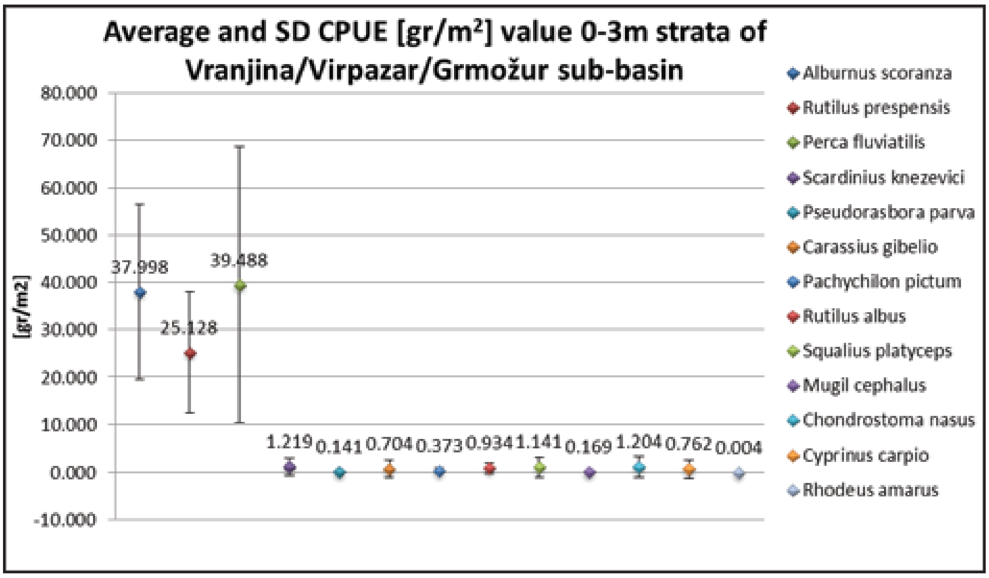


Figure 13. Average CPUE [gr/m²] and SD of CPUE [gr/m²] value 0-3m strata of Vranjina/Virpazar/Grmožur sub-basin

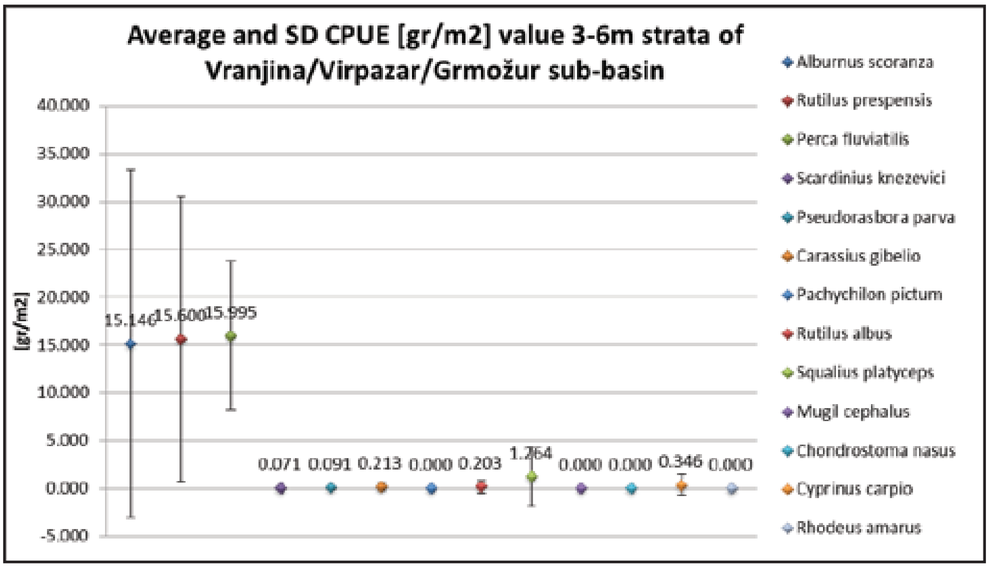
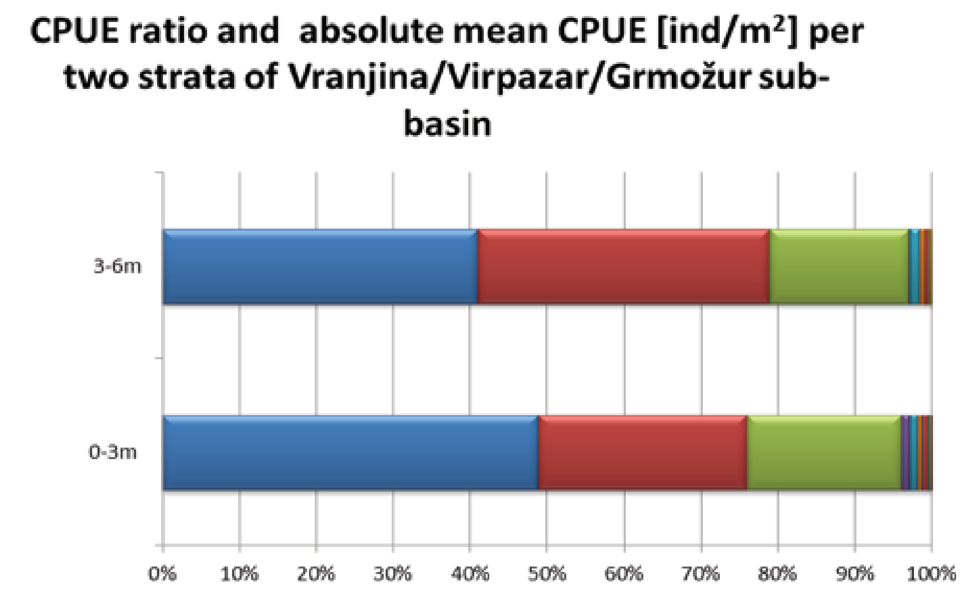


Figure 14. Average and SD CPUE [gr/m²] value 3-6m strata of Vranjina/Virpazar/ Grmožur sub- basin

Table 8. Average CPUE [gr/m²] and SD of CPUE [gr/m²] values for both strata of Vranjina/Virpazar/Grmožur sub-basin

	Alburnus scoranza	*Rutilus prespensis*	*Perca fluviatilis*	*Scardinius knezevici*	*Pseudorasbora parva*	*Carassius gibelio*	*Pachychilon pictum*	*Rutilus albus*	*Squalius platyceps*	*Mugil cephalus*	*Chondrostoma nasus*	*Cyprinus carpio*	*Rhodeus amarus*
	0-3m stratum												
Average CPUE [gr/m²]	37.998	25.128	39.488	1.219	0.141	0.704	0.373	0.934	1.141	0.169	1.204	0.762	0.004
SD	18.445	12.790	29.085	1.882	0.178	1.799	0.549	1.104	2.155	0.000	2.299	1.929	0.000
	3-6m stratum												
Average CPUE [gr/m²]	15.146	15.600	15.995	0.071	0.091	0.213	0.000	0.203	1.264	0.000	0.000	0.346	0.000
SD	18.131	14.890	7.753	0.000	0.145	0.371	0.000	0.678	3.131	0.000	0.000	1.102	0.000

Three most dominant species (bleak, perch and roach) show significantly higher absolute CPUE values in shallower 0-3m stratum. Pachychilon pictum, Mugil cephalus, Chondrostoma nasus and Rhodeus amarus were absent in deeper stratum while bigger biomass in 0-3m stratum also had Pseudorasbora parva, Scardinius knezevici and Carasius gibelio and common carp. Only chub showed higher biomass in deeper strata of the Lake (Figures 11-14 and Table 8).



	0-3m	3-6m
Alburnus scoranza	2.117	0.621
Rutilus prespensis	1.172	0.575
Perca fluviatilis	0.872	0.274
Scardinius knezevici	0.047	0.003
Pseudorasbora parva	0.039	0.019
Carassius gibelio	0.025	0.010
Pachychilon pictum	0.008	0.000
Rutilus albus	0.025	0.007
Squalius platyceps	0.006	0.004
Mugil cephalus	0.003	0.000
Chondrostoma nasus	0.006	0.000
Cyprinus carpio	0.008	0.003
Rhodeus amarus	0.003	0.000

Figure 15. CPUE ratio and absolute average CPUE [ind./m²] per two strata of Vranjina/Virpazar/Grmožur sub-basin

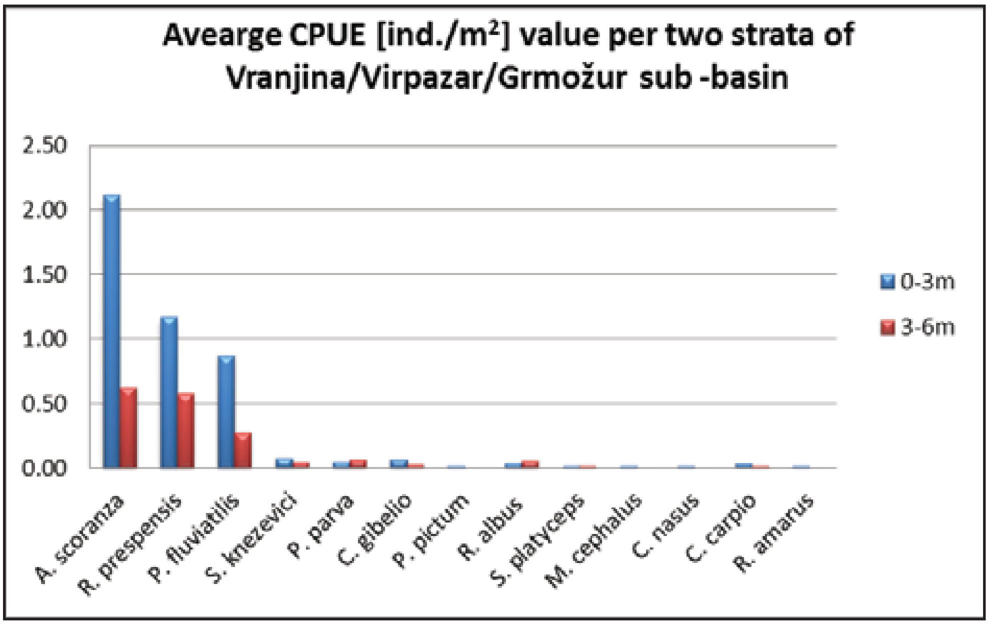


Figure 16. Comparative review of calculated average CPUE values (ind./m² of MMG net) for all sampled species in both strata

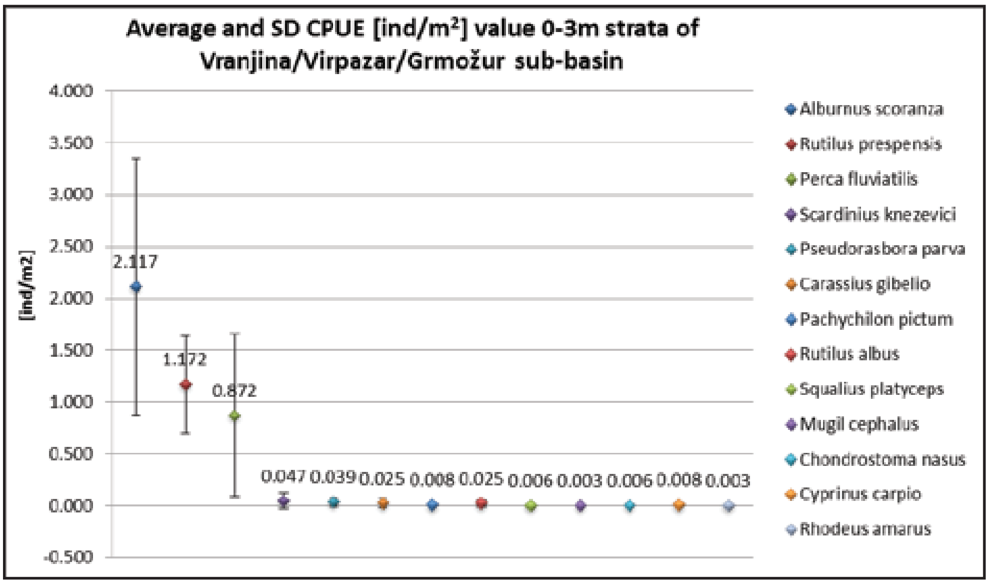


Figure 17. Average CPUE [ind./m²] and SD of CPUE [ind./m²] values in 0-3m strata of Vranjina/Virpazar/Grmožur sub-basin

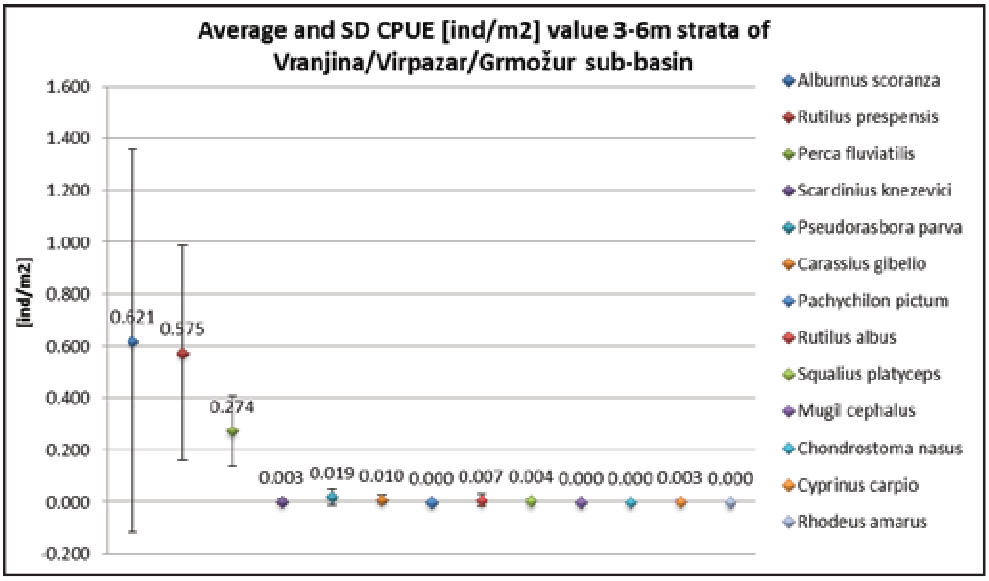


Figure 18. Average CPUE [ind./m²] and SD of CPUE [ind./m²] values in 3-6m strata of Vranjina/Virpazar/Grmožur sub-basin

Table 9. Average CPUE [ind./m^2] and SD of CPUE [ind/m^2] values for both strata of Vranjina/Virpazar/Grmožur sub-basin

	Alburnus scoranza	*Rutilus prespensis*	*Perca fluviatilis*	*Scardinius knezevici*	*Pseudorasbora parva*	*Carassius gibelio*	*Pachychilon pictum*	*Rutilus albus*	*Squalius platyceps*	*Mugil cephalus*	*Chondrostoma nasus*	*Cyprinus carpio*	*Rhodeus amarus*
	0-3m stratum												
Average CPUE [ind./m^2]	2.117	1.172	0.872	0.047	0.039	0.025	0.008	0.025	0.006	0.003	0.006	0.008	0.003
SD	1.235	0.470	0.784	0.076	0.044	0.047	0.012	0.030	0.010	0.000	0.010	0.017	0.000
	3-6m stratum												
Average CPUE [ind/m^2]	0.621	0.575	0.274	0.003	0.019	0.010	0.000	0.007	0.004	0.000	0.000	0.003	0.000
SD	0.737	0.414	0.133	0.011	0.030	0.016	0.000	0.022	0.009	0.000	0.000	0.007	0.000

Considering CPUE values in terms of numbers of individuals per one m^2 of MMG net we can see similar tendency. Here we have again situation that CPUE value for bleak, perch and roach is higher in 0-3m stratum but now almost three times, suggesting bigger size fish in deeper waters. *Scardinius knezevici* and *Carassius gibelio* shows double higher CPUE in shallower part of the Lake (silver carp has higer SD in shallower part) while common carp, chub, *Pseudorasbora parva* and *Rutilus albus* had almost the same CPUE values regarding number of the individuals (Figure 15-18 and Table 9).

4.3.1.1.3. Population structure

Based on MMG gillnet data sets (0-3 m and 3-6 m strata) we can analyze populations of the three most abundant species (bleak, perch and roach) that also has highest CPUE values. Other species individuals were not present in such numbers in this gear samples, not even in both strata pulled, so it was not possible to do following analysis for theme.

Alburnus scoranza (bleak)

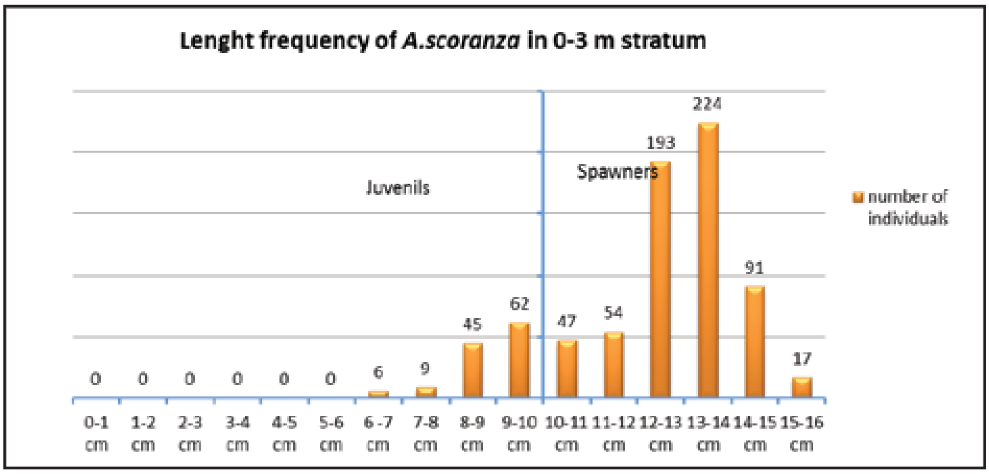


Figure 19. Length frequency of bleak (Alburnus scoranza) in 0-3m stratum

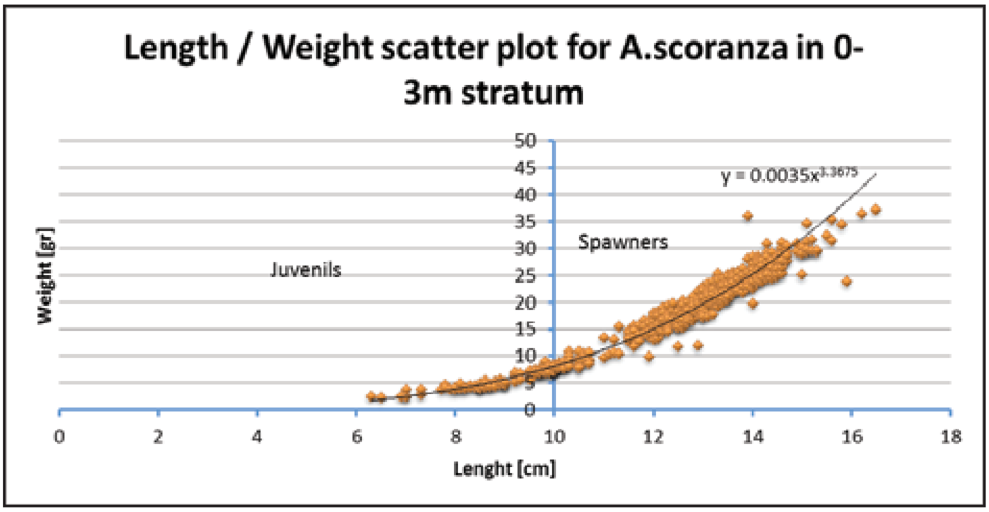


Figure 20. Length/Weight scatter plot for bleak (Alburnus scoranza) in 0-3 m stratum

In part of bleak population from 0-3m strata we have sampled fish from all length intervals with dominance of 13-14 cm and 12-13 cm length class. In this strata we had significant number of juveniles but spawners were far more dominant (Figure 19. and 20). Those data indicate presence of all age cohorts in this Lake zone (Figure 19 and 20.).

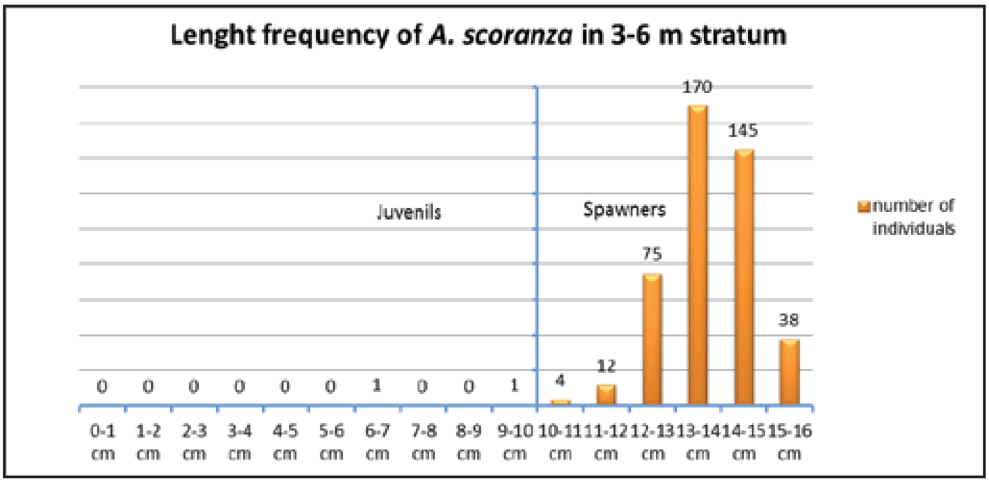


Figure 21. Length frequency of bleak (Alburnus scoranza) in 3-6m stratum

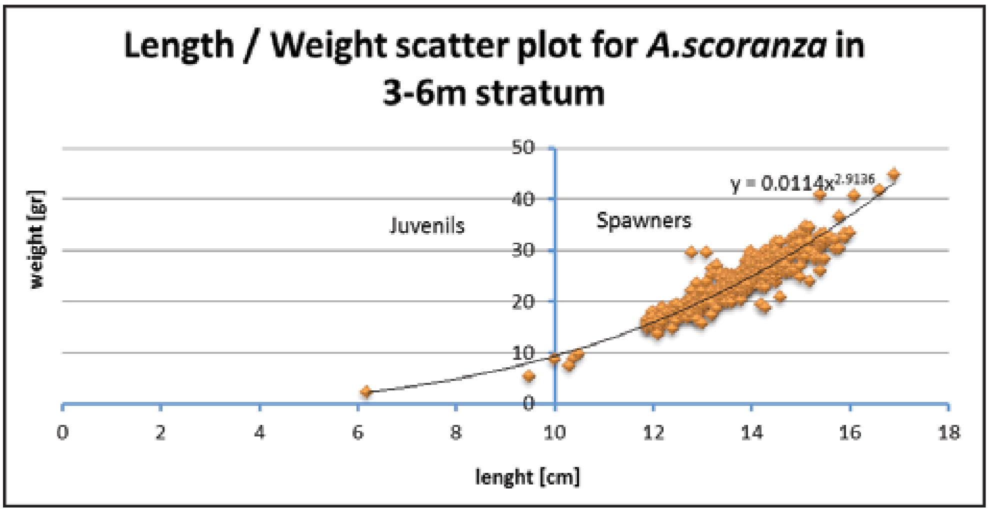


Figure 22. Length/Weight scatter plot for bleak (Alburnus scoranza) in 3-6 m stratum

In part of bleak population from 3-6m strata we haven't sampled fish from all length intervals while dominate were those of 13-14 cm and 14-15 cm length class. This graph indicates presence of older age cohorts in this Lake zone while younger were almost absent (only one individual of 6-7cm length class and one from 9-10 class) (Figure 21 and 22.). Such situation suggests that, in deep Lake area in this part of the year, it occurs mainly adult-spawners individuals of bleak population.

Perca fluviatilis (perch)

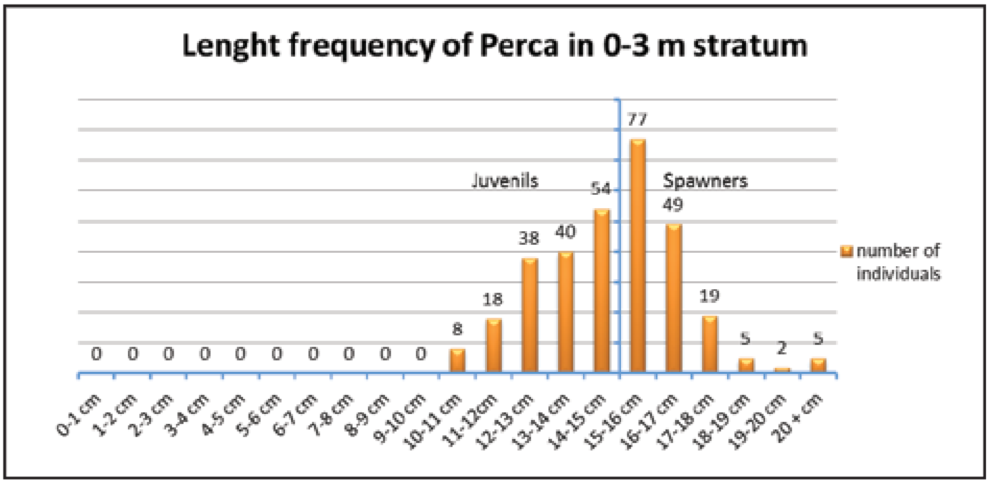


Figure 23. Length frequency of perch (P. fluviatilis) in 0-3m stratum

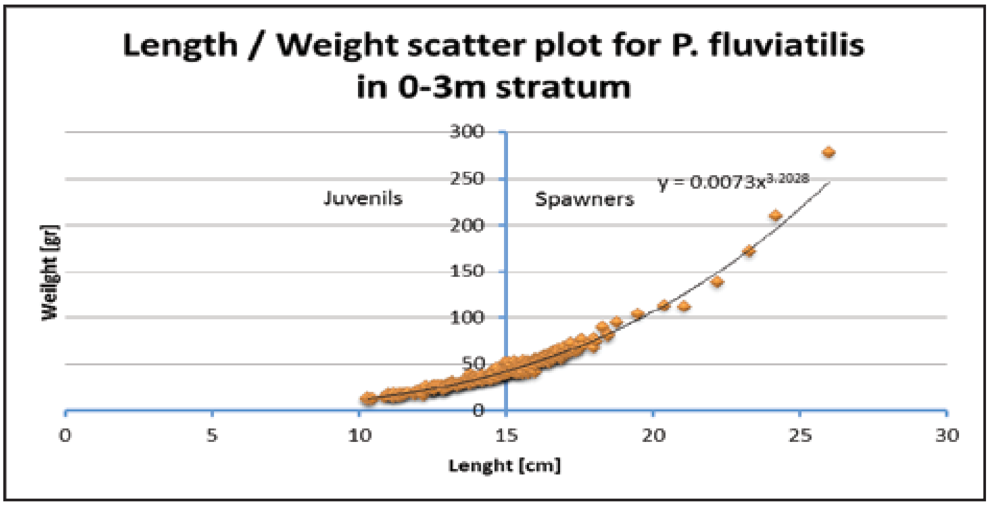


Figure 24. Length/Weight scatter plot for perch (P. fluviatilis) in 0-3 m stratum

In part of perch population from 0-3m strata we have sampled fish from all length intervals with dominance of 15-16cm, 14-15cm and 16-17cm length classes respectively (Figure 23). This graph indicates presence of all age cohorts in this Lake zone but hire dominant were juveniles (Figure 23. and 24.).

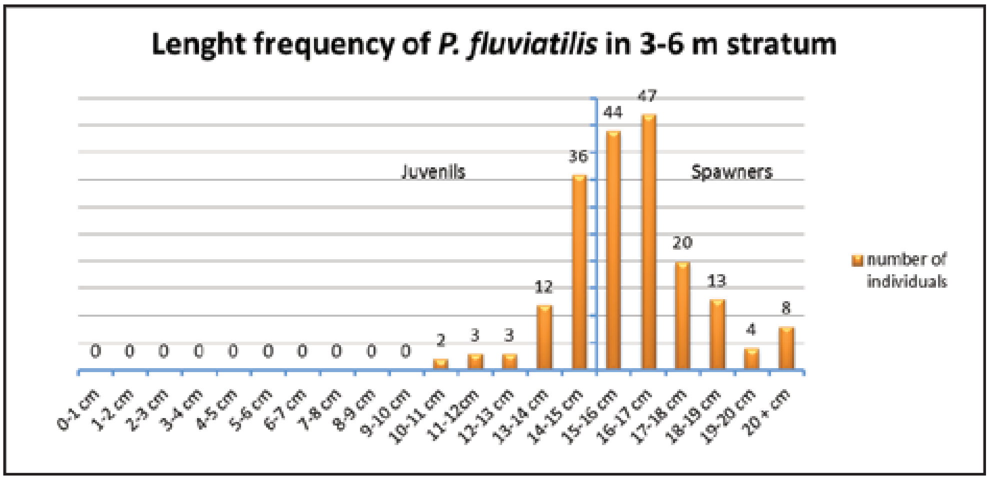


Figure 25. Length frequency of perch (P. fluviatilis) in 3-6m stratum

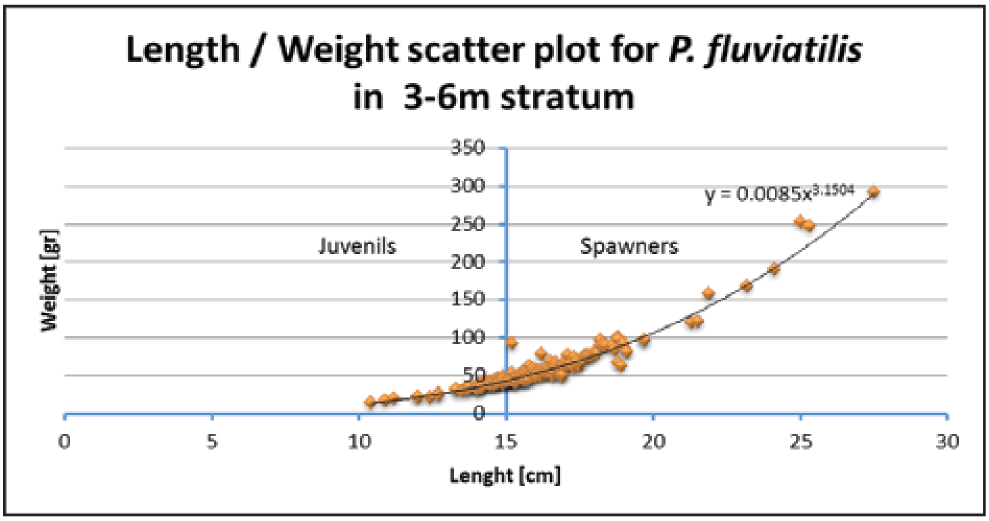


Figure 26. Length/Weight scatter plot for perch (P. fluviatilis) in 3-6 m stratum

In 3-6m strata dominant were 16-17cm, 15-16cm and 14-15cm length class, respectively (Figures 25. and 26.). Comparing of length frequencies in 3-6m strata with those in 0-3m strata we can notice that perch population in deeper water masses is consisting of slight larger individuals and juveniles are not dominant as in shallower zone of the lake (Figures 25 and 26). In this zone there are present of all age classes also but in different proportions compared to 0-3m stratum (Figures 25. and 26.).

Rutilus prespensis (roach)

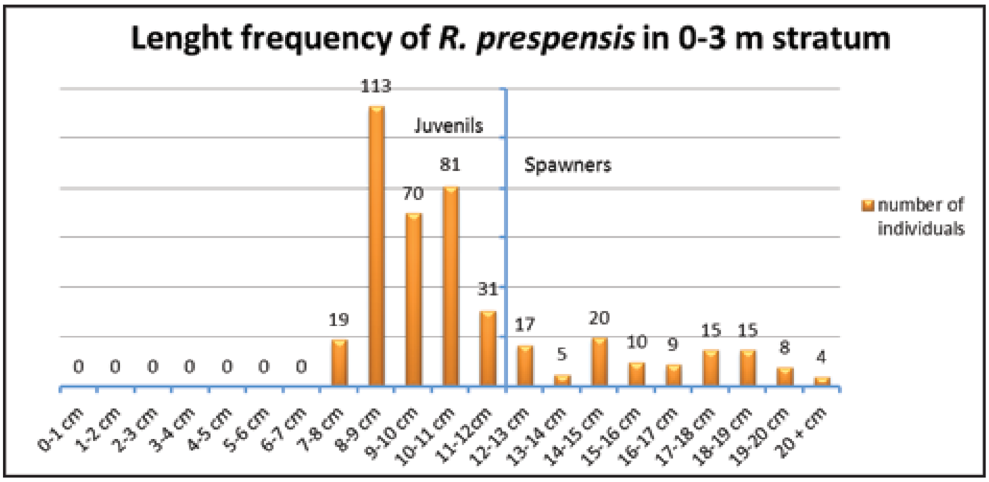


Figure 27. Length frequency of roach (R. prespensis) in 0-3m stratum

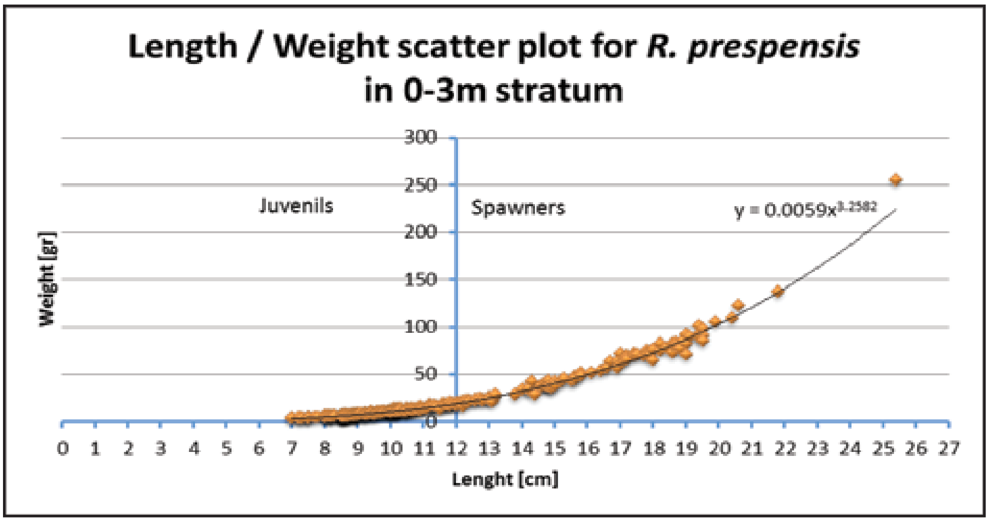


Figure 28. Length/Weight scatter plot for perch (R. prespensis) in 0-3 m stratum

In part of roach population from 0-3m strata we have sampled fish from all length intervals with dominance of 8-9cm, 10-11cm and 9-10cm length classes respectively. Those graphs indicate presence of all age cohorts in this Lake zone with dominance of juvenile ones (Figures 27 and 28).

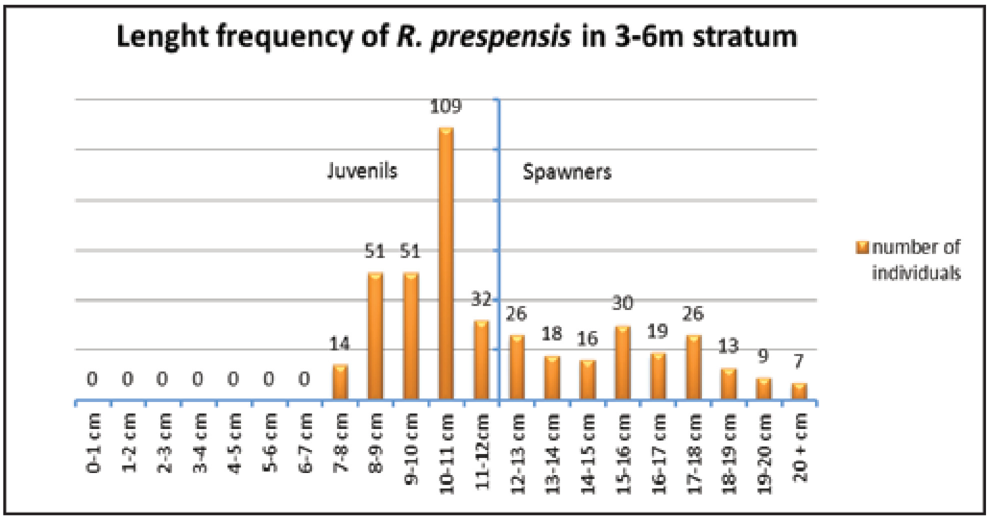


Figure 29. Length frequency of roach (R. prespensis) in 3-6m stratum

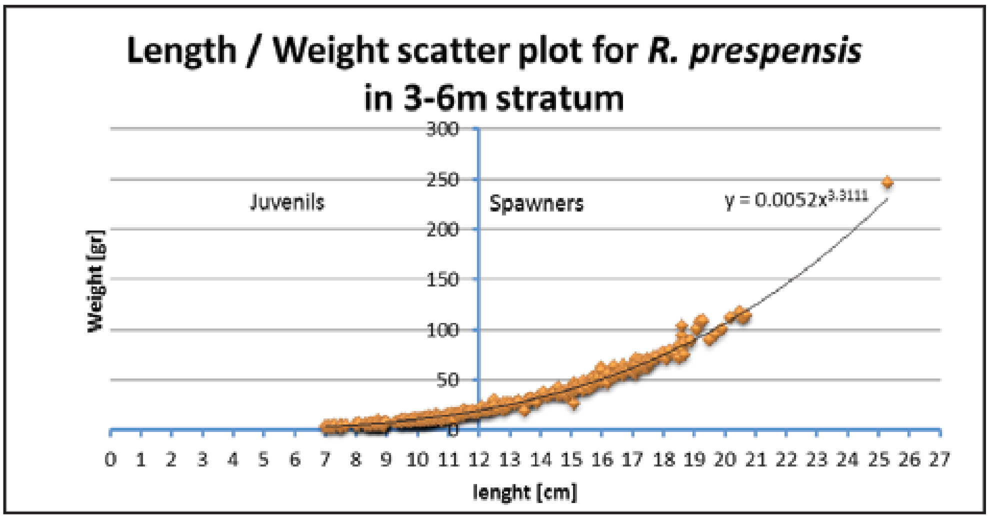


Figure 30. Length/Weight scatter plot for perch (R. prespensis) in 3-6 m stratum

In 3-6 m strata dominant were 10-11cm, 8-9 cm and 9-10 cm length class, respectively (Figure 17.). Comparing of length frequencies in 3-6 m strata with those in 0-3 m strata we can notice that roach population in deeper water masses is consisting of slight larger individuals from older age class (Figures 16. and 17.). In contrast to shallow water masses, length class of 15-16 cm and 17-18 cm has higher frequency in deeper zone of Lake suggesting less dominance of juvenile individuals in 3-6m strata.

4.3.1.2 Electro-fishing transects

In all three transects we detect in total 9 species with slight difference among individual transects (Figure 25.). All three transects were performed in 0-3m stratum.

4.3.1.2.1. Diversity and CPUE values

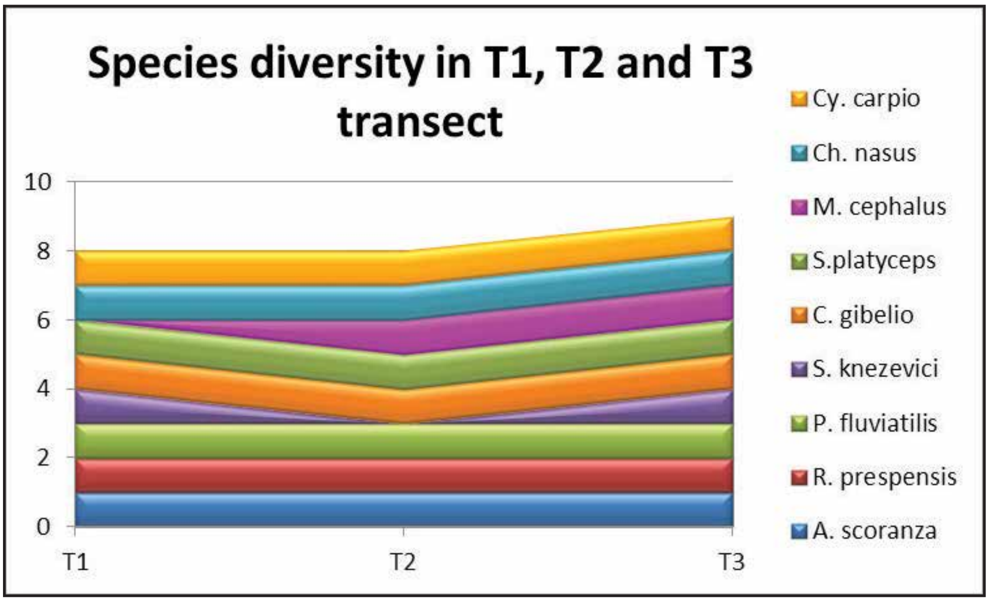
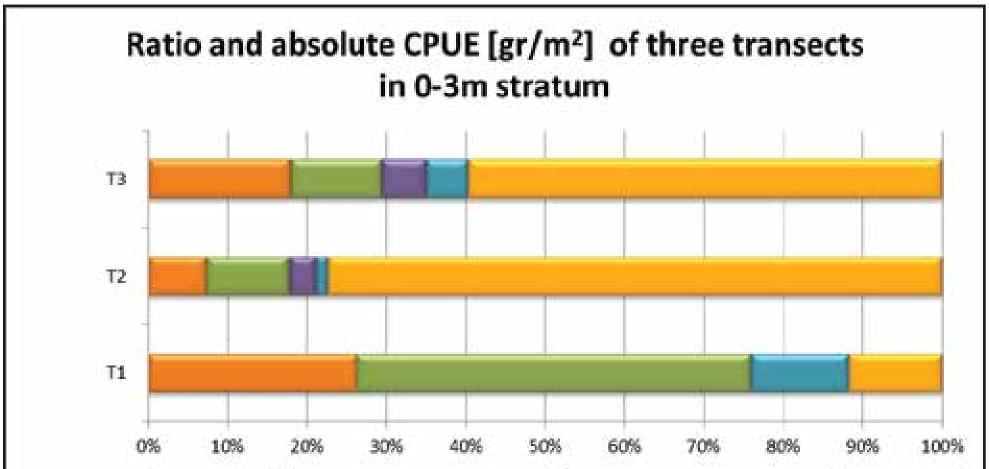


Figure 31. Species diversity in electro fishing transects, presence-absence (T1, T2 and T3)

CPUE values were calculated for surface of 2000 m² (each transect was 500m long and 4m wide). Considering that MMG gillnets show less efficiency in netting of fish with larger body and contrary for CPUE calculation we chose following five of theme that were almost absent in MMG nets samples (Figure 32.).



	T1	T2	T3
C. gibelio	1.6974	0.87995	0.8201
S.platyceps	3.2141	1.23955	0.52105
M. cephalus		0.41395	0.25245
Ch. nasus	0.80085	0.1572	0.24755
Cy. carpio	0.76145	9.22865	2.7239

Figure 32. Ratio and absolute CPUE [gr/m²] in all three transects for five fish species (T1, T2 and T3)

Analyzing of CPUE value proportion among all three transects we can conclude that for two transects (T2 and T3) we had dominance of carp (Cyprinus carpio) while chub (Squalius platyceps) was dominant in third transect (T1).

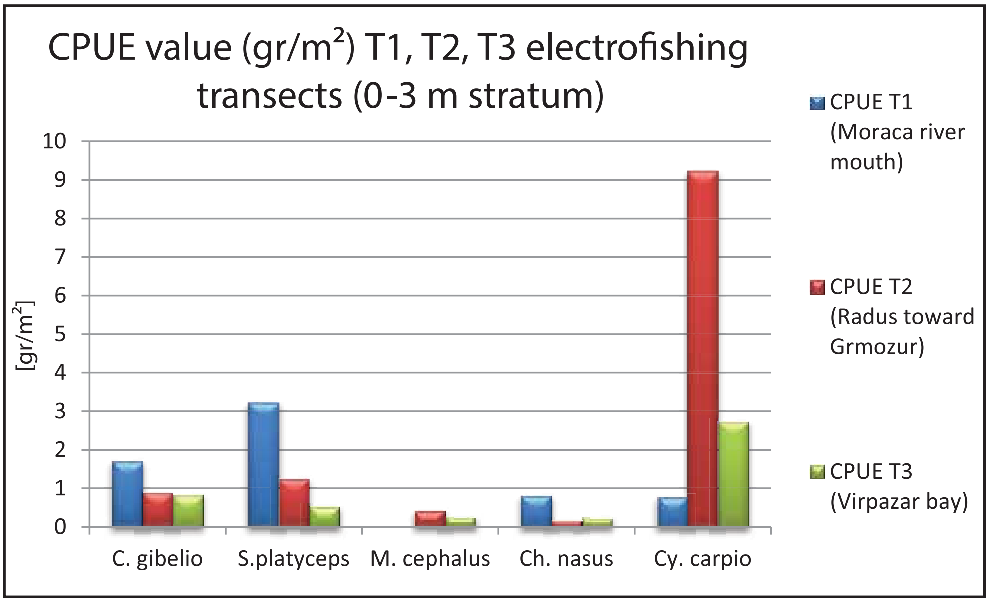
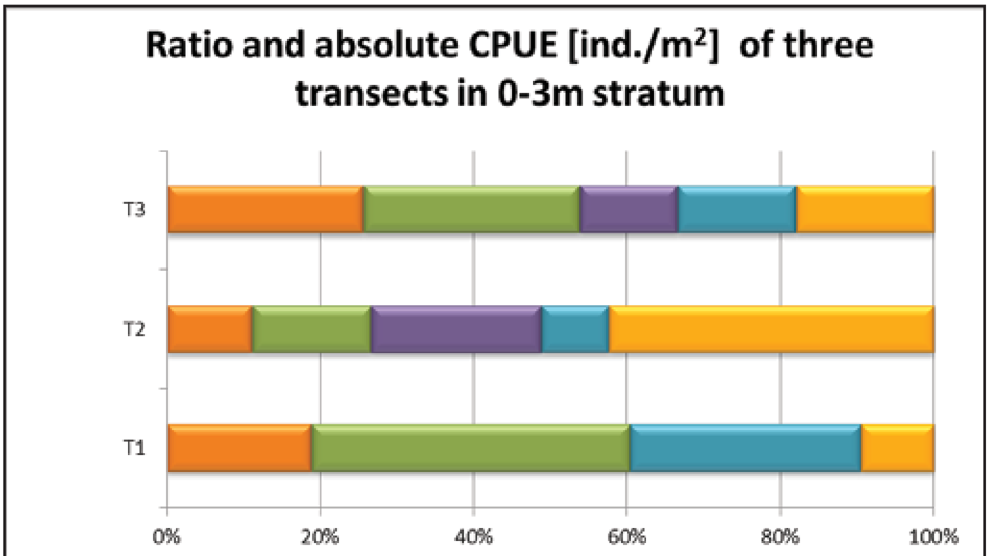


Figure 33. Absolute CPUE [gr/m²] values in all three transects (T1, T2 and T3)

In T1 transect highest CPUE value has chub (S. platyceps) while in T3 transect highest CPUE value showed carp (carp and chub had similar CPUE value in those two locations). In T3 transect dominant was carp and CPUE value was by far highest among all species in all tree transects.

Table 10. Absolute number of individuals per transect (T1, T2 and T3)

	C.gibelio	*S.platyceps*	*M.cephalus*	*Ch. nasus*	*Cy.carpio*
T1	10	22		16	5
T2	5	7	10	4	19
T3	10	11	5	6	7



	T1	T2	T3
C.gibelio	0.005	0.003	0.005
S.platiceps	0.011	0.004	0.006
M.cephalus	0.000	0.005	0.003
Ch. nasus	0.008	0.002	0.003
Cy.carpio	0.003	0.010	0.004

Figure 34. Ratio and absolute CPUE [ind./m²] in all three transects for five fish species (T1, T2 and T3)

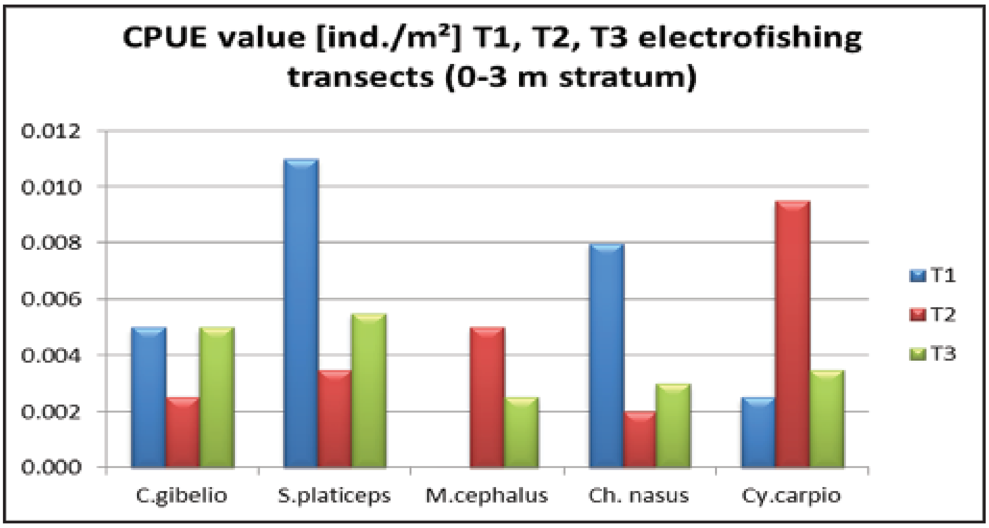


Figure 35. Absolute CPUE [ind./m^2] values in all three transects (T1, T2 and T3)

Chub shows highest abundance in transects T1 and T2 while common carp have highest abundance in T3 transects. Chub and nase shows higher population density in transect near Morača river inlet while common carp prefer typical Lake habitats with rocky bottom (Raduš-Grmožur trisect T3) (Figure 34 and 35).

4.3.1.3 Commercial bleak harvesting (Raduš bay)

Following data and graphs are based on subsample which we take from the total "kalimera" net catch.

4.3.1.3.1. Diversity in commercial catch

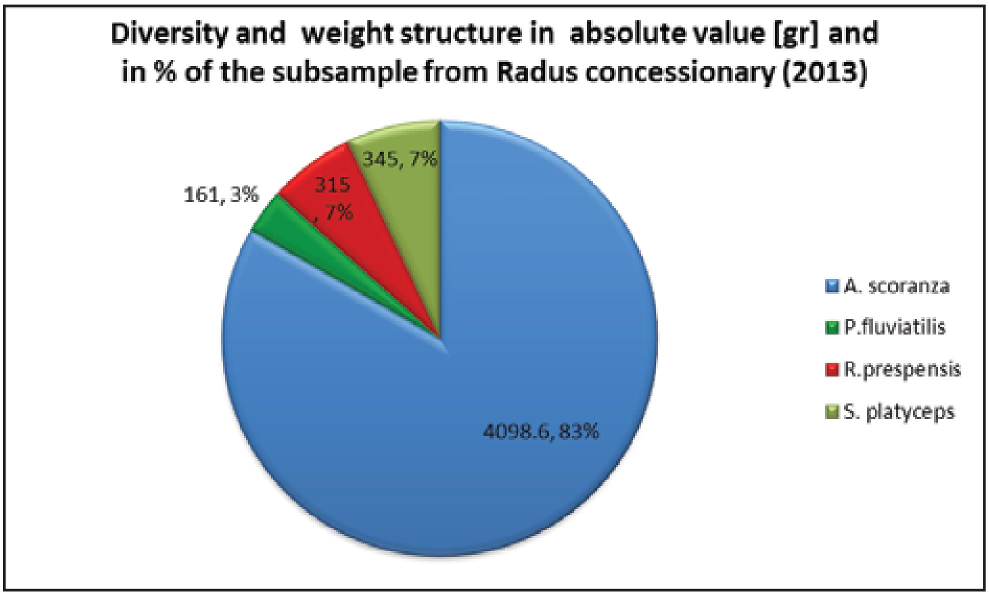


Figure 36. Diversity and weight structure of the catch from Raduš concessionary (2013) (absolute number, percentage)

Harvesting season occurs during wintering season when bleak forms several huge shoals in Lake bays, and one of theme is Raduš where bleak forms the most dense and biggest shoal in whole lake. So it is not surprising that bleak is far more dominant in harvesting catch (by mass and by abundance). Other accompanied species that takes the same habitats during winter haven't shows some significant biomass in 2013th sampling campaign.

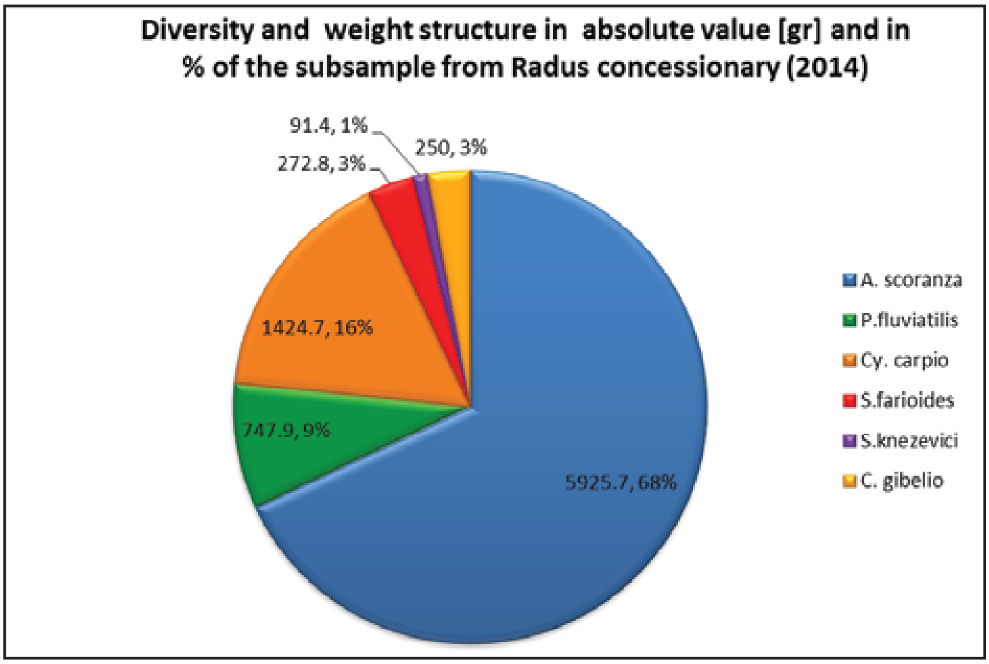


Figure 37. Diversity and weight structure of the catch from Raduš concessionary (2014)

In 2014th structure of the "kalimera" catch was significantly different comparing to 2013th samples. Namely in 2014th proportion of bleak was significantly lower than in 2013th and catch was characterized with higher species diversity (6 species). Beside significant amount of carp in 2014th subsample we also had brown trout in it (morpha lacustris of Salmo farioides). Presence of brown trout could indicate overall good conditions of Lake fish fauna as well as good condition of whole lake ecosystem.

4.3.1.3.2. Population structure of bleak (Alburnus scoranza) from Raduš concessionary

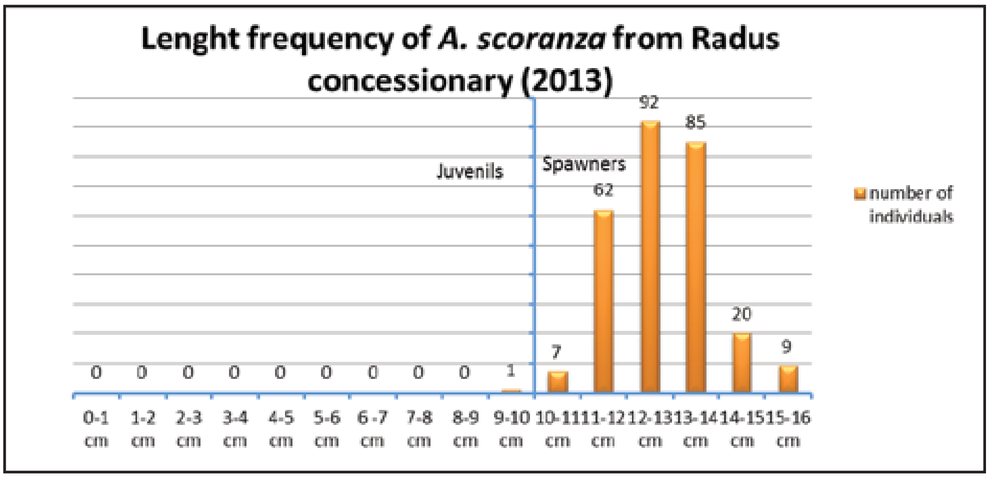


Figure 38. Length frequency of bleak (A. scoranza) from Raduš concessionary (2013)

In subsample taken from "kalimera" catch in 2013th dominant were 12-13cm, 13-14cm and 11-12cm length classes, respectively. In graph it is obvious that in catch sample we are missing younger age class (smaller length classes, Figure 38).

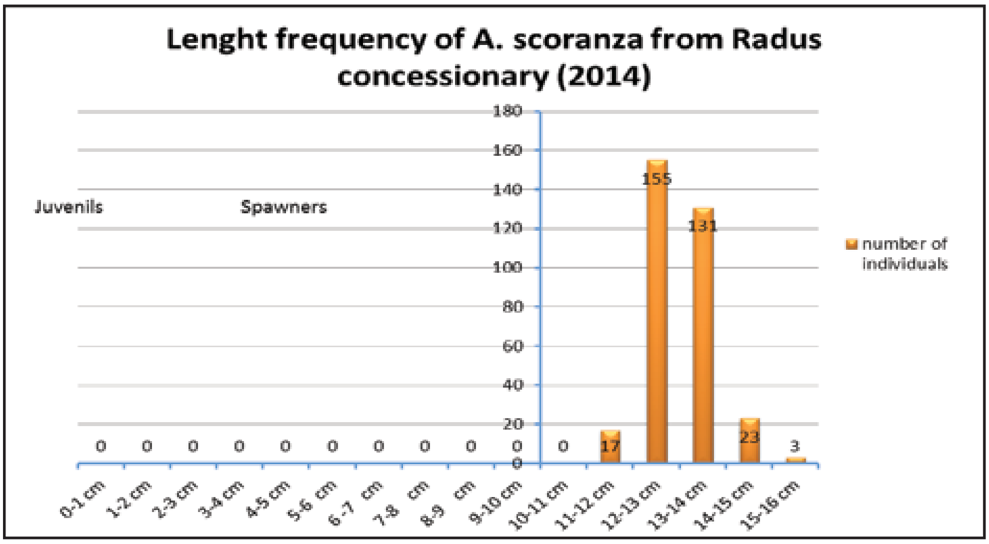


Figure 39. Length frequency of bleak (A. scoranza) from Raduš concessionary (2014)

In subsample taken from "kalimera" catch in 2014th dominant were 12-13cm and 13-14cm length classes, respectively. In graph it is obvious that in catch sample there are no younger age classes (6-11 cm length class) (Figure 39.).

4.3.2. Country: Albania

4.3.2.1 Multimesh gillnetting

4.3.2.1.1. Abundance

Table 11. Number of each species individual caught in both sub-basins

Name of species	Shirokë		Koplik	
	No. of ind.	Max. size [cm]	No. of ind.	Max. size [cm]
Alburnoides ohridanus	15	9	6	11
Alburnus scoranza	651	17	238	17
Allosa fallax	-	-	34	15
Carassius gibelio	9	17	1	9
Chondrostoma nasus	46	12	125	10
Cobitis meridionalis	2	8	3	8
Cyprinus carpio	18	25	6	13
Pachychilon pictum	155	15	74	13
Perca fluviatilis	11	20	38	22
Pseudorasbora parva	380	10	303	10
Rhodeus amarus	81	7	62	7
Rutilus rubilio	582	17	718	19
Scardinus erythrophthalmus	34	25	5	21
Total	**1984**		**1613**	

Koplik area

Although 11 species were caught in Koplik, there is a large dominance of Rutilus, Alburnus and Pseudorasbora that represent nearly 80 % of all individuals (Figures 40, 41 and 42). Rutilus is more present in 0-3 and 3-6 meters depth, while in more than 6 meters depth, Rutilus is less abundant. The opposite situation is with Alburnus which is less abundant in the first two strata and more abundant in the last one. Pseudorasbora presence is constant in all three strata.

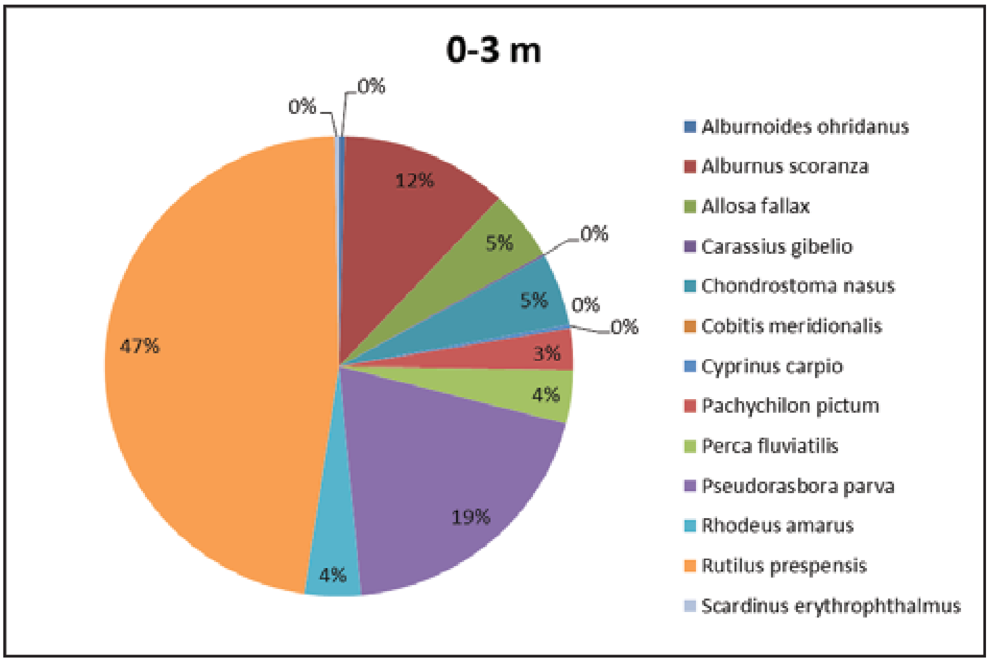


Figure 40. Abundance of species in 0-3m stratum catch (Koplik sub-basin)

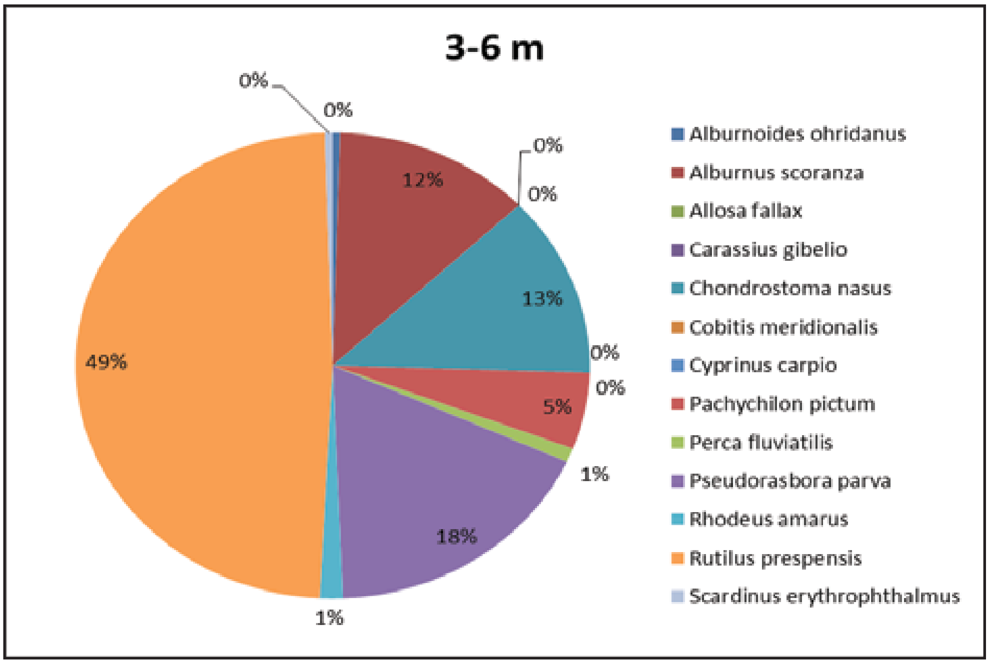


Figure 41. Abundance of species in 3-6m stratum catch (Koplik sub-basin)

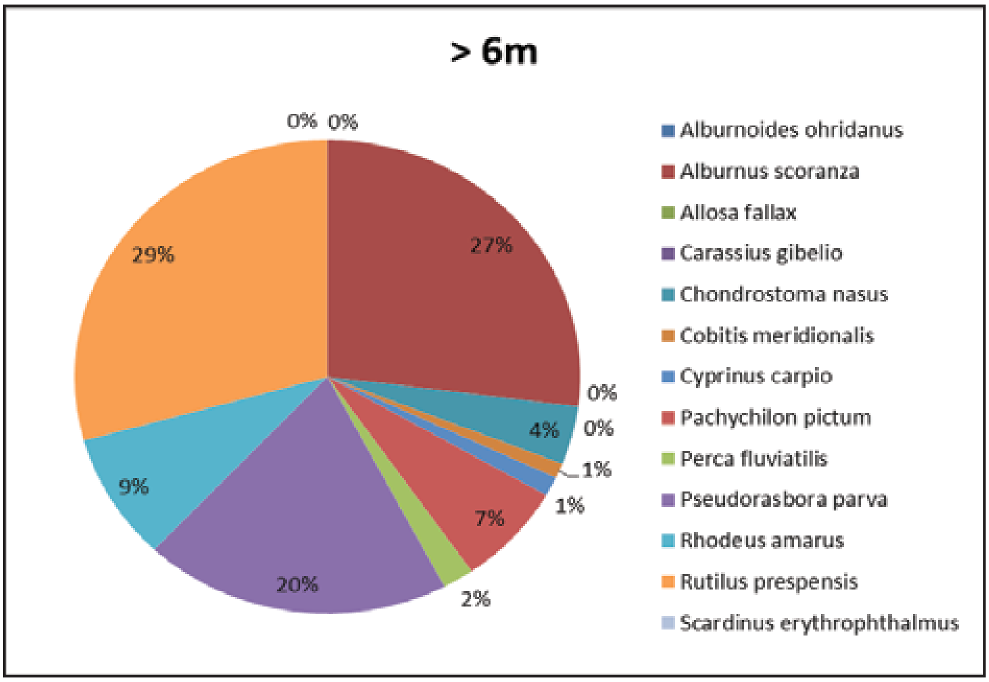


Figure 42. Abundance of species in >6m stratum catch (Koplik sub-basin)

Shirokë area

Although 11 species were caught in Shirokë, there is a large dominance of Rutilus, Alburnus and Pseudorasbora that represent nearly 75 % of all individuals (Figures 42, 43 and 44). Rutilus is more present in 0-3 depth, while 3-6 meters and in more than 6 meters depth, Rutilus is less abundant. In Shiroka we should note than Alburnus is more abundant than Rutilus compared with Koplik. The opposite situation is with Alburnus which is less abundant in the first two strata and more abundant in the last one. Pseudorasbora presence is notable in all three strata although it is less abundant in the 0-3 meters strata.

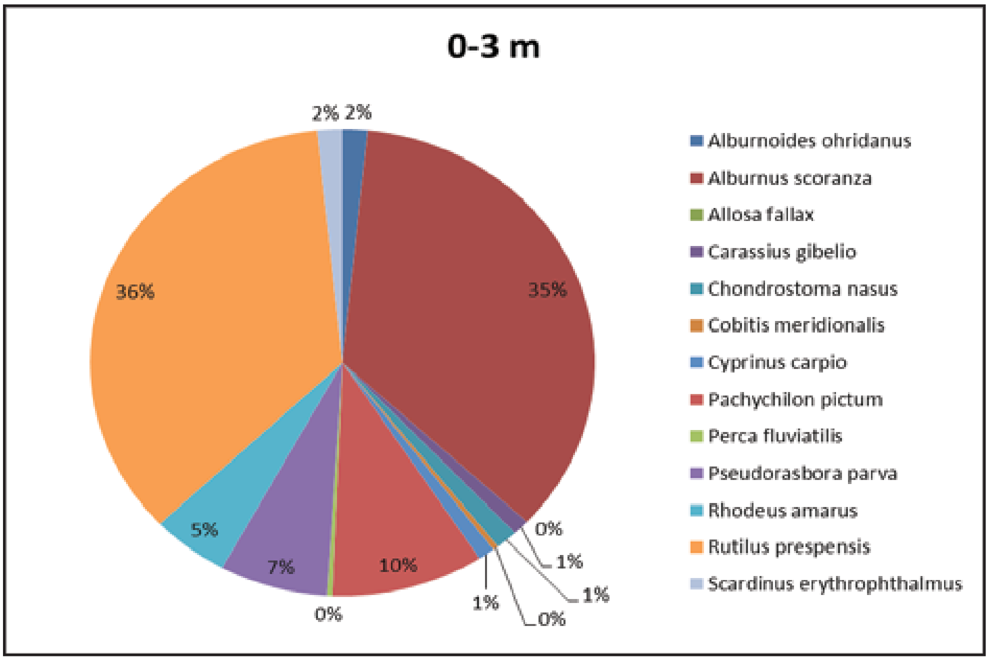


Figure 43. Abundance of species in 0-3m stratum catch (Shirokë sub-basin)

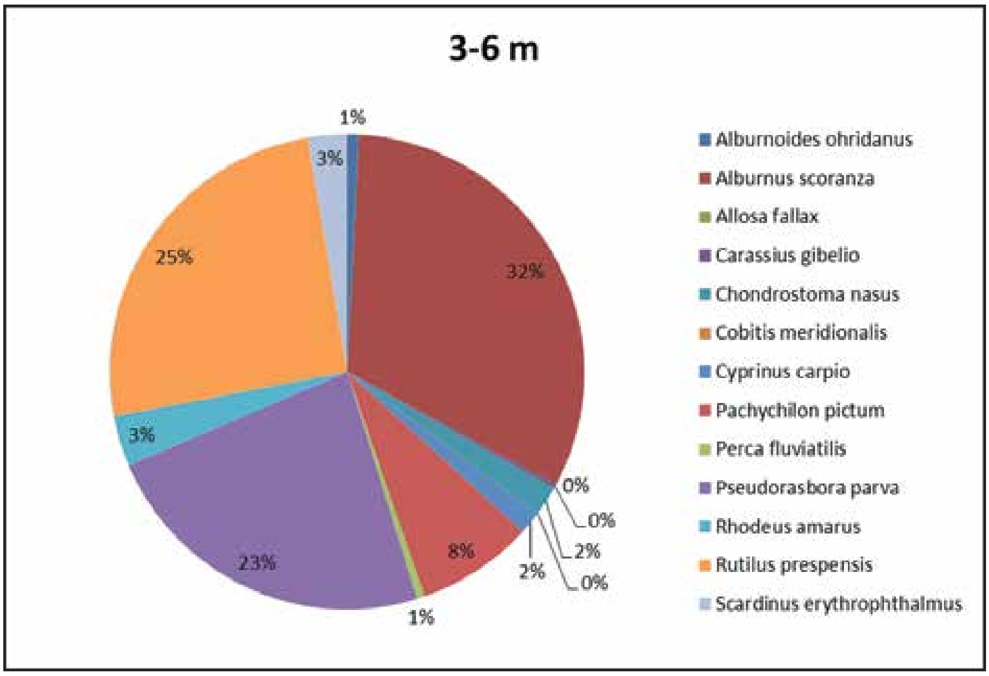


Figure 44. Abundance of species in 3-6m stratum catch (Shirokë sub-basin)

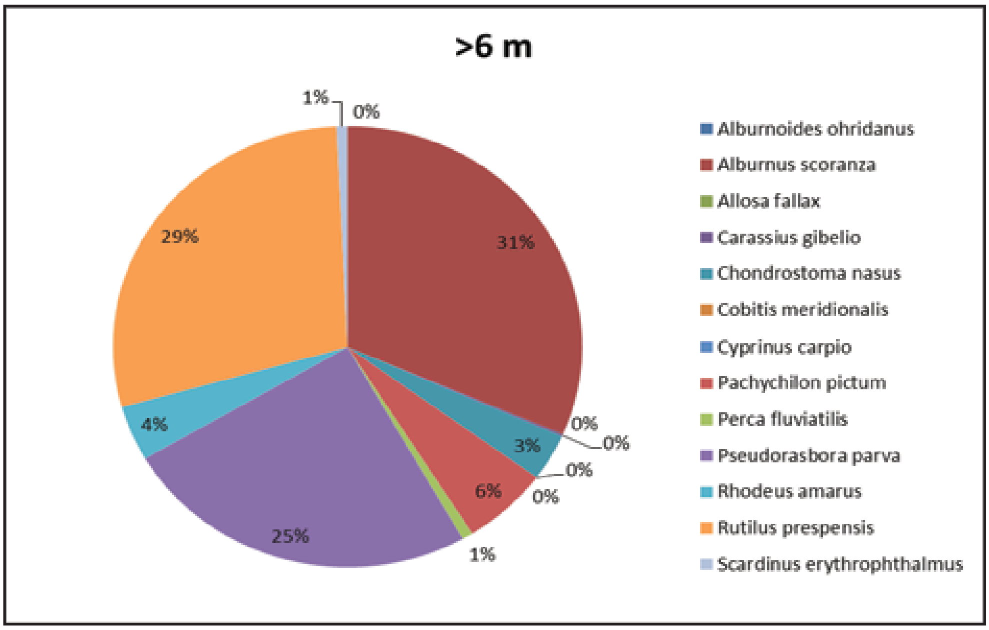
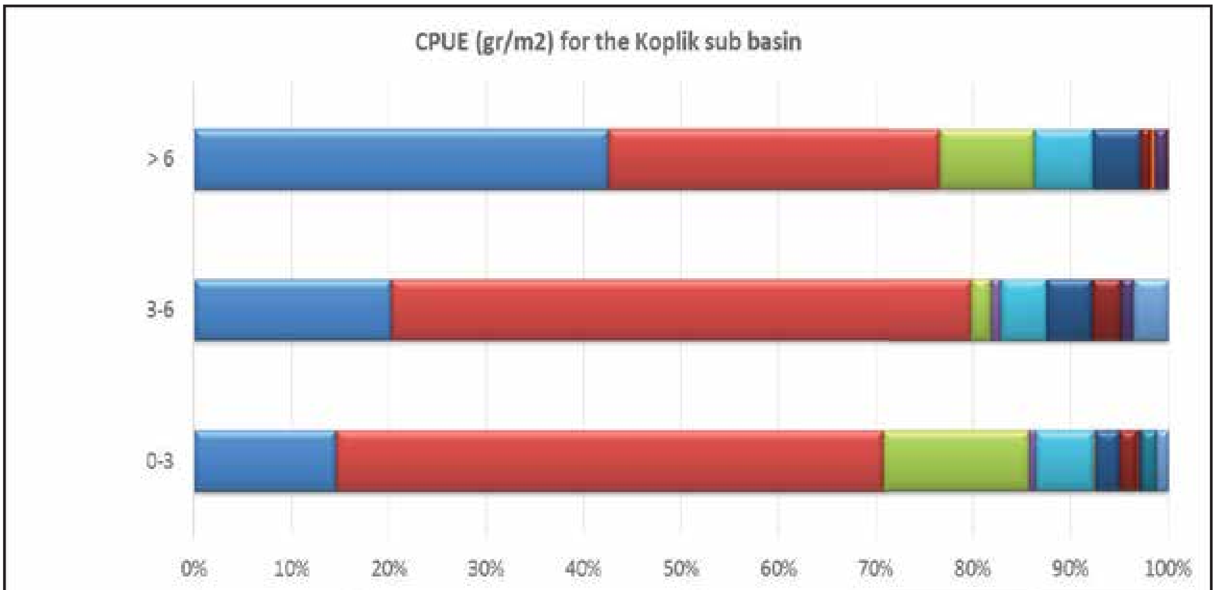


Figure 45. Abundance of species in >6m stratum catch (Shirokë sub-basin)

4.3.2.1.2. CPUE

Koplik area

Following figure shows CPUE values ratio for all sampled species per all three strata (Figure 46.). In Koplik area we found that CPUE of bleak rise with depth while for roach it is just opposite. Those two species together with perch in >6m stratum represent more than 85 % of total catch, in 3-6 stratum they are more than 81 % while in shallowest stratum (0-3m of depth) they represent nearly 86 %. Regarding biomass as well as abundance roach is most common species in total catch 0-3 and 3-6m strata while bleak is dominant in deepest stratum (>6m of depth).



	0-3	3-6	> 6
Alburnus scoranza	1.630666667	2.646851852	3.334814815
Rutilus prespensis	6.280888889	7.832592593	2.674074074
Perca fluviatilis	1.689777778	0.261481481	0.758518519
Scardinus knezevici	0.072592593	0.138888889	
Pseudorasbora parva	0.661777778	0.609074074	0.473333333
Carassius gibelio	0.019555556		
Pachychilon pictum	0.272148148	0.610555556	0.378024691
Chondrostoma nasus	0.204148148	0.391666667	0.082222222
Cyprinus carpio	0.016296296		0.03382716
Rhodeus amarus	0.036	0.162222222	0.098765432
Allosa fallax	0.176888889		
Cobitis meridionalis			0.010123457
Alburnoides ohridanus	0.131407407	0.47037037	

Figure 46. CPUE ratio and absolute average CPUE [gr/m²] per three strata of Koplik sub-basin

In following figure there are shown absolute CPUE values for four most abundant and most important (in terms of weight of total catch) fish species (Figure 47.)

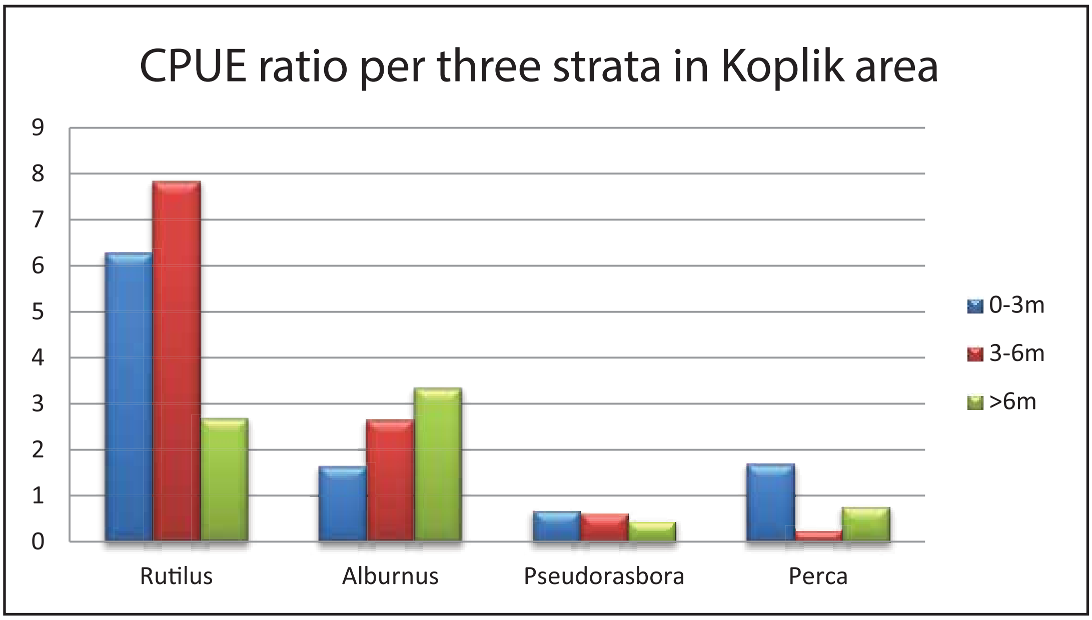


Figure 47. Comparative review of calculated average CPUE values (gr/m² of MMG net) for four major fish species in all three strata of Koplik sub-basin

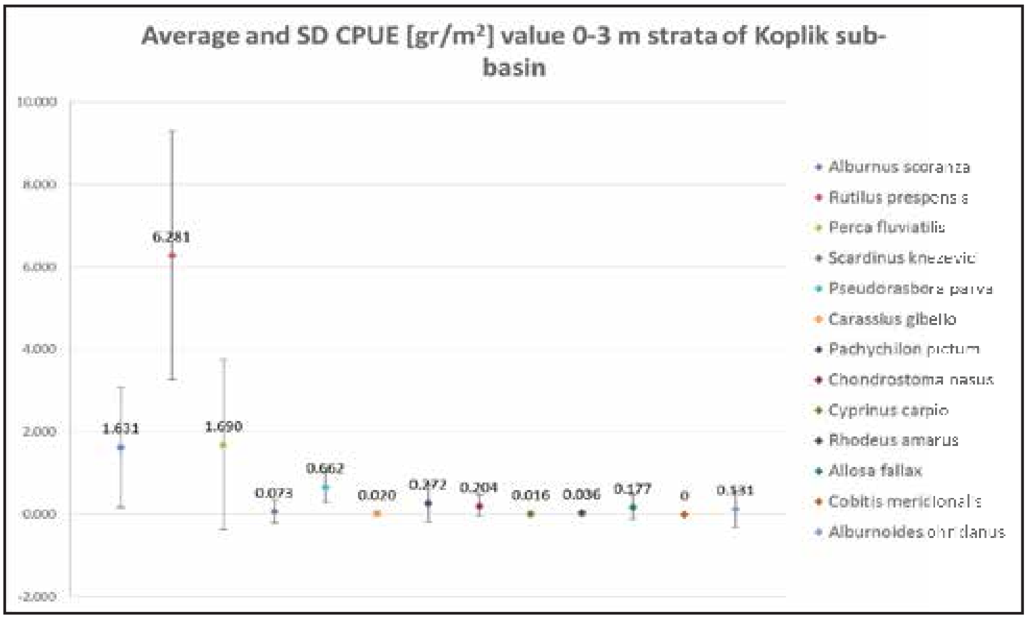


Figure 48. Average CPUE [gr/m²] and SD of CPUE [gr/m²] value 0-3m strata of Koplik sub-basin

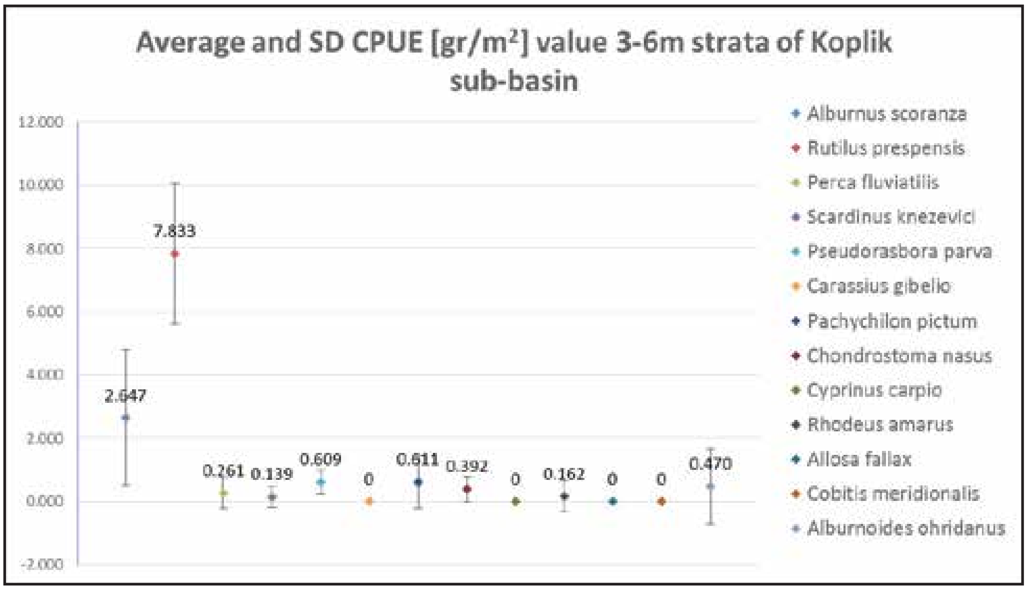


Figure 49. Average CPUE [gr/m²] and SD of CPUE [gr/m²] value 3-6m strata of Koplik sub-basin

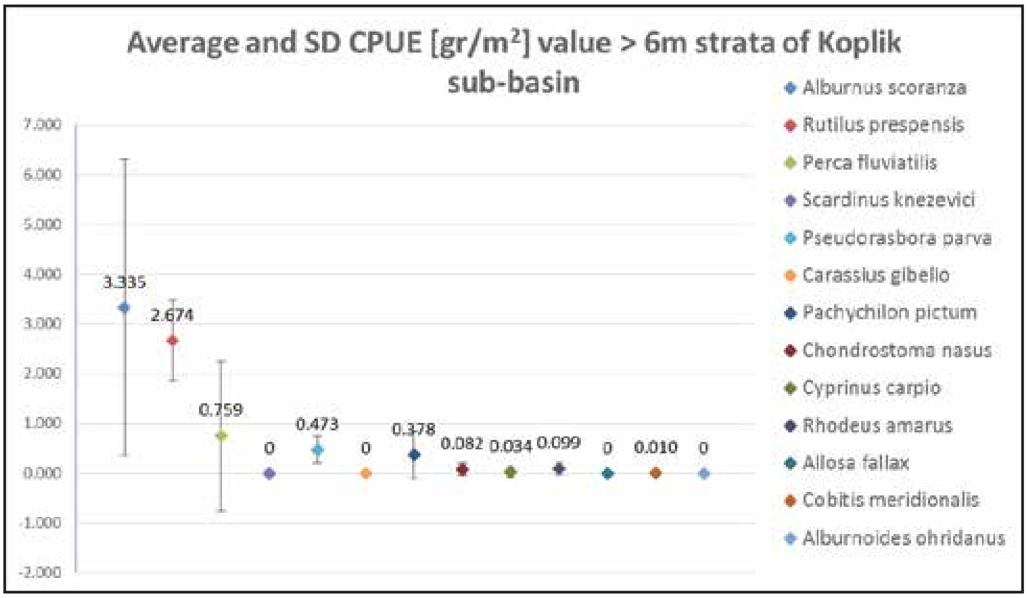


Figure 50. Average CPUE [gr/m²] and SD of CPUE [gr/m²] value >6m strata of Koplik sub-basin

Table 12. Average CPUE [gr/m^2] and SD of CPUE [gr/m^2] values for three strata of Koplik sub-basin

	0-3m		3-6m		>6m	
	Average CPUE [gr/m2]	SD	Average CPUE [gr/m2]	SD	Average CPUE [gr/ m2]	SD
Alburnus scoranza	1.631	1.458	2.647	2.144	3.335	2.978
Rutilus prespensis	6.281	3.010	7.833	2.229	2.674	0.814
Perca fluviatilis	1.690	2.051	0.261	0.493	0.759	1.507
Scardinus knezevici	0.073	0.281	0.139	0.342	0.000	0.000
Pseudorasbora parva	0.662	0.375	0.609	0.380	0.473	0.269
Carassius gibelio	0.020	0.076	0.000	0.000	0.000	0.000
Pachychilon pictum	0.272	0.462	0.611	0.839	0.378	0.481
Chondrostoma nasus	0.204	0.258	0.392	0.390	0.082	0.125
Cyprinus carpio	0.016	0.063	0.000	0.000	0.034	0.101
Rhodeus amarus	0.036	0.061	0.162	0.493	0.099	0.110
Allosa fallax	0.177	0.296	0.000	0.000	0.000	0.000
Cobitis meridionalis	0.000	0.000	0.000	0.000	0.010	0.021
Alburnoides ohridanus	0.131	0.447	0.470	1.185	0.000	0.000

As it is shown on Table 12, the fish biomass is more abundant in the 0-3 and 3-6 m depth while in the > 6m, the biomass tends to decrease for all species except bleak.

In following graphs and table we are presenting CPUE in terms of numbers of individuals per one m^2 of MMG net. Those graphs are reflecting relative abundance of sampled species in Koplik sub-basin.

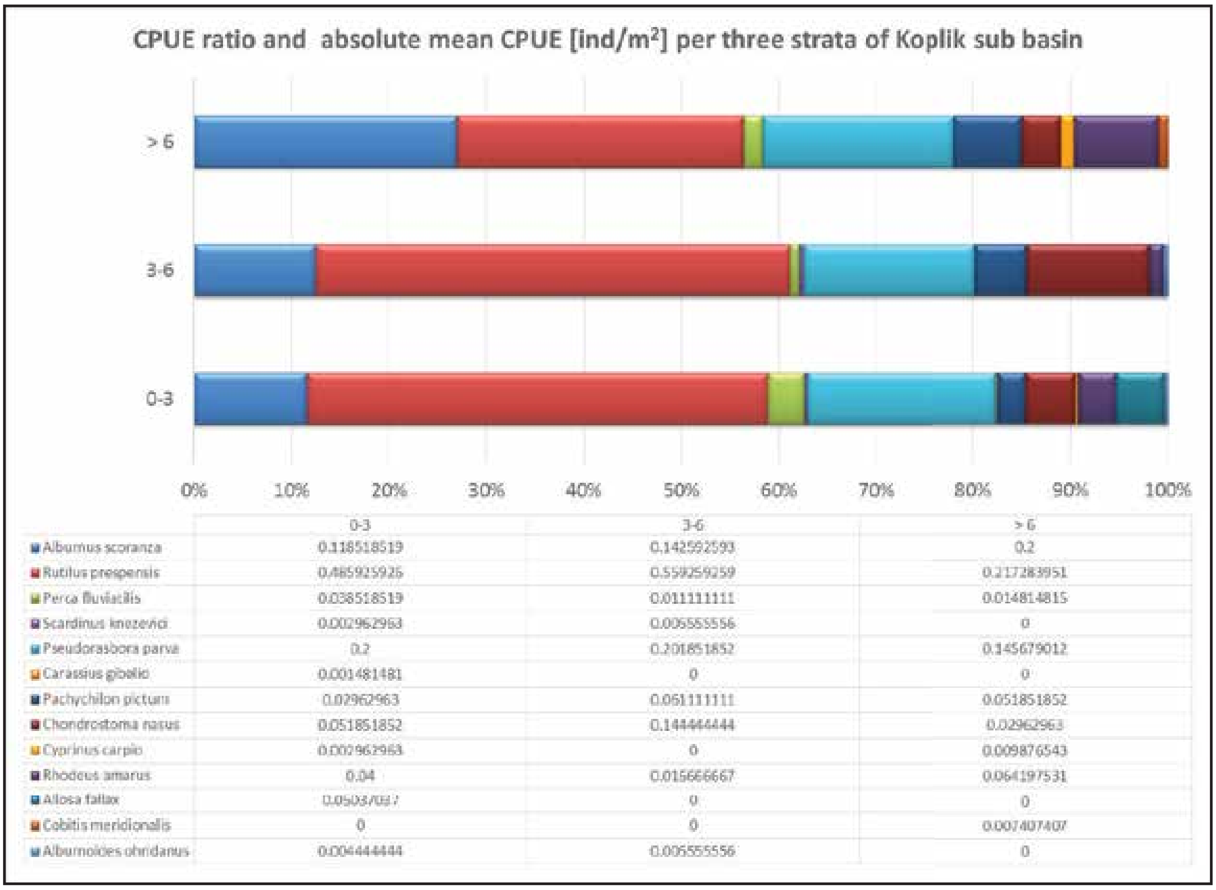


Figure 51. CPUE ratio and absolute average CPUE [ind/m^2] per three strata of Koplik sub-basin

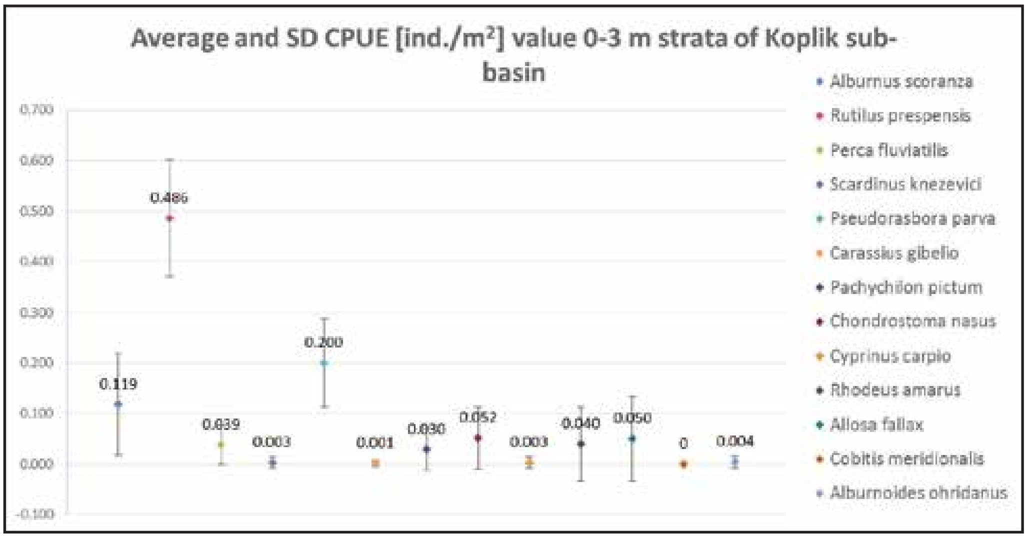


Figure 52. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value 0-3m strata of Koplik sub-basin

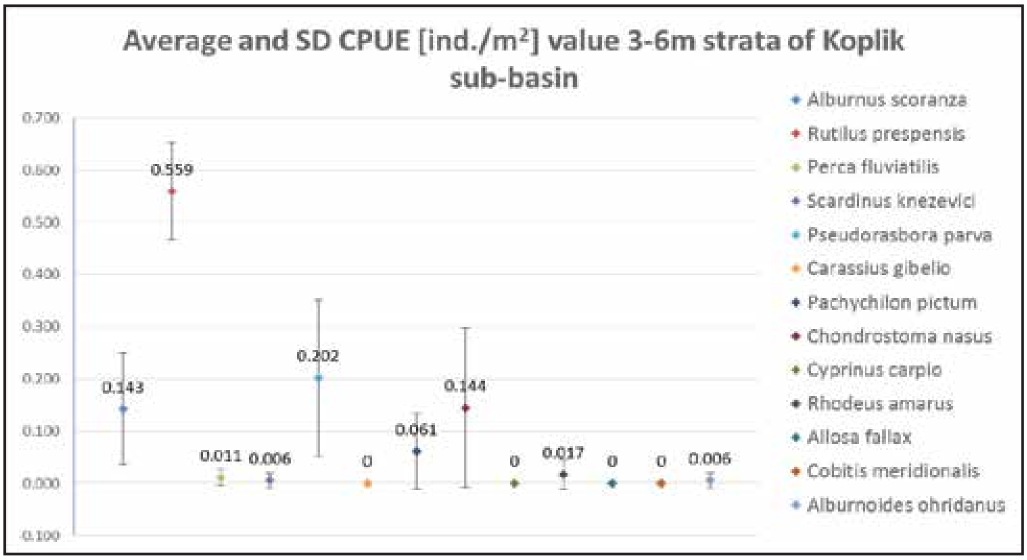


Figure 53. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value 3-6m strata of Koplik sub-basin

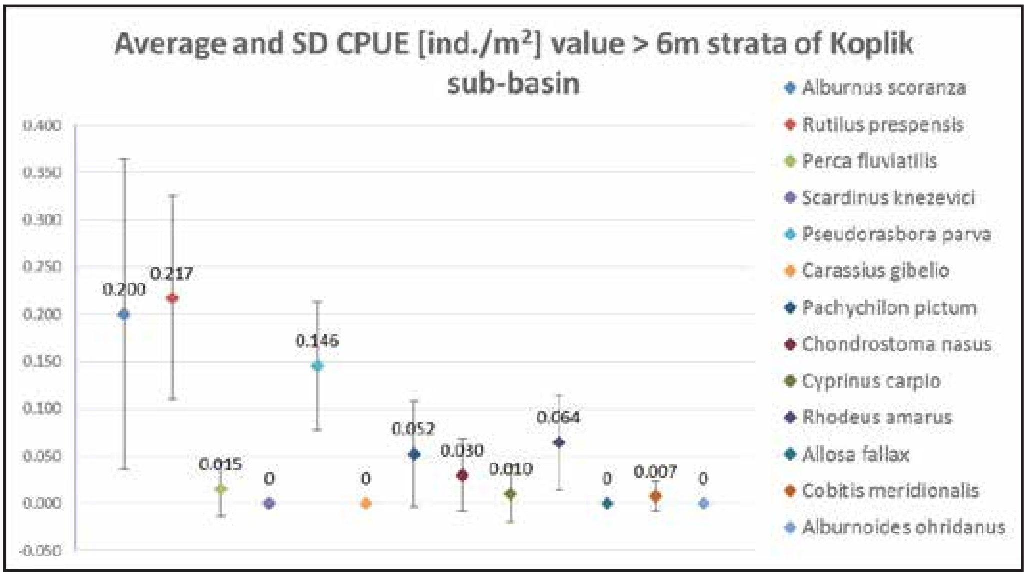


Figure 54. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value >6m strata of Koplik sub-basin

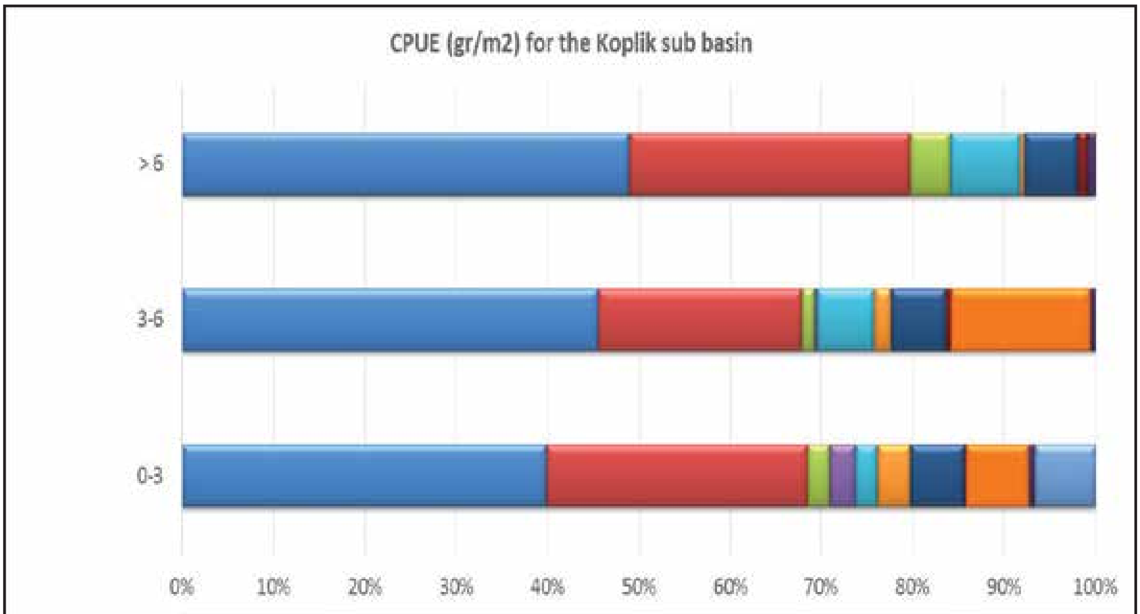
Table 13. Average CPUE [ind/m²] and SD of CPUE [ind/m²] values for three strata of Koplik sub-basin

	0-3m		3-6m		>6m	
	Average CPUE [ind./m2]	SD	Average CPUE [ind./m2]	SD	Average CPUE [ind./m2]	SD
Alburnus scoranza	0.119	0.101	0.143	0.107	0.200	0.164
Rutilus prespensis	0.486	0.115	0.559	0.092	0.217	0.108
Perca fluviatilis	0.039	0.039	0.011	0.015	0.015	0.029
Scardinus knezevici	0.003	0.011	0.006	0.014	0.000	0.000
Pseudorasbora parva	0.200	0.086	0.202	0.150	0.146	0.068
Carassius gibelio	0.001	0.006	0.000	0.000	0.000	0.000
Pachychilon pictum	0.030	0.043	0.061	0.073	0.052	0.056
Chondrostoma nasus	0.052	0.061	0.144	0.152	0.030	0.038
Cyprinus carpio	0.003	0.011	0.000	0.000	0.010	0.030
Rhodeus amarus	0.040	0.073	0.017	0.029	0.064	0.050
Allosa fallax	0.050	0.083	0.000	0.000	0.000	0.000
Cobitis meridionalis	0.000	0.000	0.000	0.000	0.007	0.016
Alburnoides ohridanus	0.004	0.012	0.006	0.014	0.000	0.000

As it is shown, in Table 13, when we reevaluate the CPUE from the number of individuals, the dominance of small size species is more emphasized and the values of large size species such as P. fluviatilis and S. knezevici are very small. However, the trend of lower biomass in the > 6m depth compared to shallower waters is evident also when we consider the number of individuals.

Shirokë area

Following figure shows CPUE values ratio for all sampled species per all three strata (Figure 55.). In contrast to Koplik area here we have bleak as most dominant species in terms of total catch (gr). In Shiroka area, bleak also shows rising of total biomass with rising of strata depth while for roach there is no such or opposite trend (Figure 55.). In this area, carp also appears as significant in total catch but only in middle strata (3-6m). This is consequence of bigger body size of this fish species and if we analyze graph for abundance (Figures 43-45.) we can notice that carp is almost irrelevant in terms of numbers of individuals. Comparing of CPUE ratio from Shiroka with the same from Koplik area we can also conclude that in this region perch is far less abundant (in terms of numbers of individuals as well as in terms of total biomass). In Shiroka, the species Pachychilon pictum plays significant role instead of perch (in terms of total biomass of catch)



	0-3	3-6	> 6
Alburnus scoranza	4.269	8.419	6.474
Rutilus prespensis	3.074	4.155	4.091
Perca fluviatilis	0.255	0.254	0.576
Scardinus knezevici	0.306	0.055	0.013
Pseudorasbora parva	0.255	1.147	1.006
Carassius gibelio	0.381	0.339	0.059
Pachychilon pictum	0.636	1.112	0.772
Chondrostoma nasus	0.023	0.098	0.145
Cyprinus carpio	0.728	2.810	
Rhodeus amarus	0.065	0.088	0.112
Allosa fallax			
Cobitis meridionalis	0.003		
Alburnoides ohridanus	0.716	0.027	

Figure 55. CPUE ratio and absolute average CPUE [gr/m²] per three strata of Shirokë sub-basin

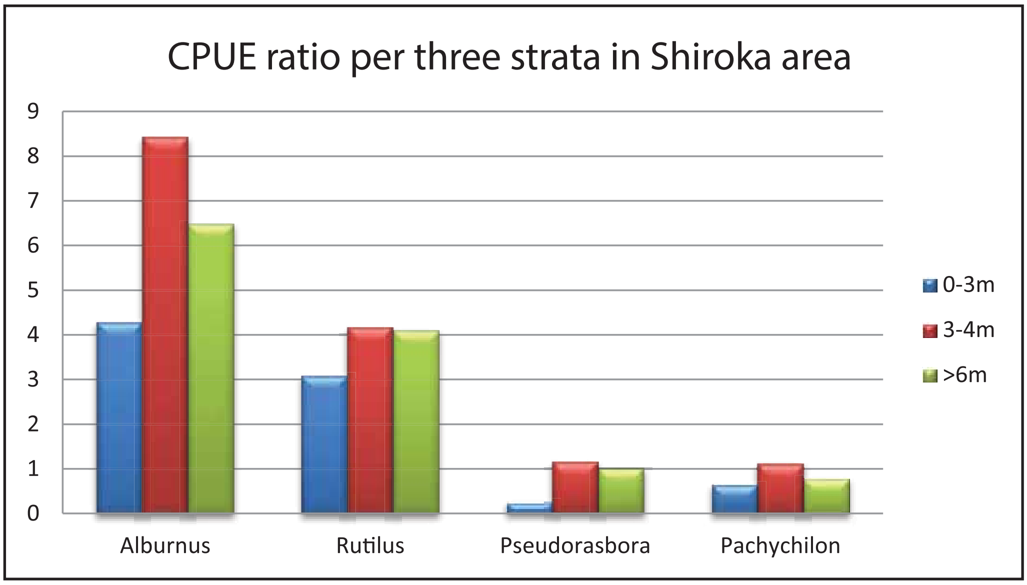


Figure 56. Comparative review of calculated average CPUE values (gr/m² of MMG net) for four major fish species in all three strata of Shirokë sub-basin

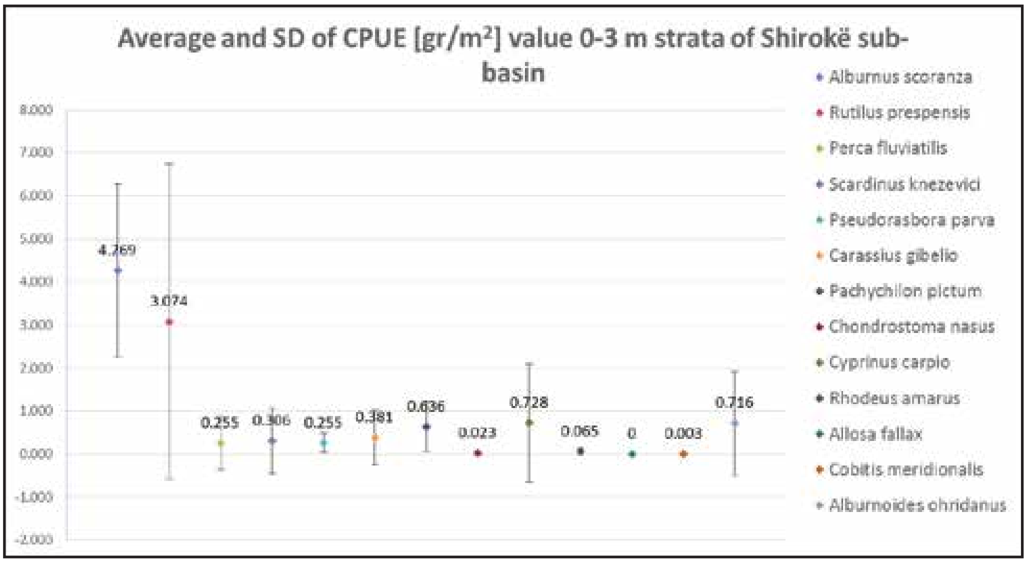


Figure 57. Average CPUE [gr/m^2] and SD of CPUE [gr/m^2] value 0-3m strata of Shirokë sub-basin

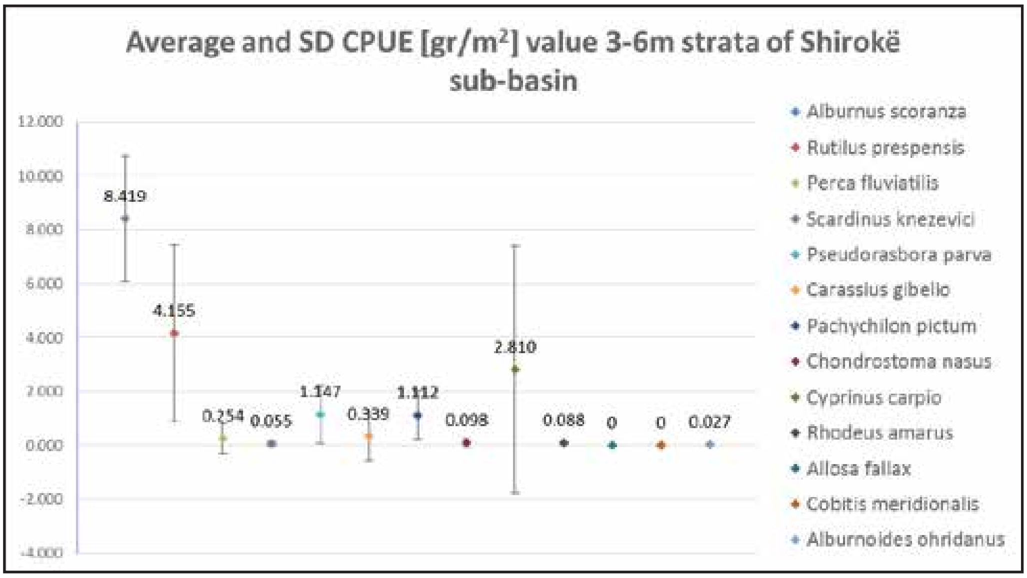


Figure 58. Average CPUE [gr/m^2] and SD of CPUE [gr/m^2] value 3-6m strata of Shirokë sub-basin

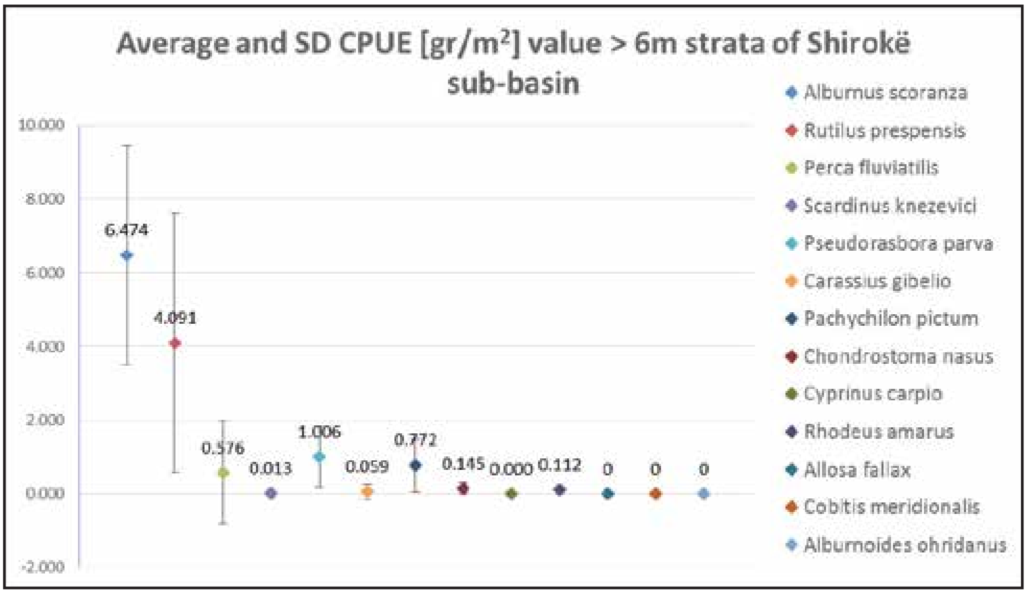


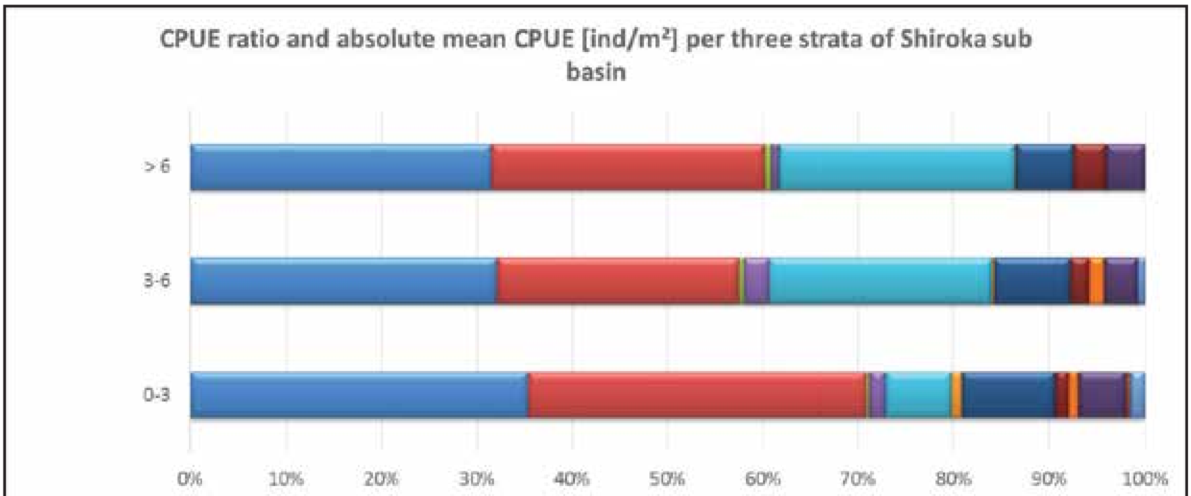
Figure 59. Average CPUE [gr/m^2] and SD of CPUE [gr/m^2] value >6m strata of Shirokë sub-basin

Table 14. Average CPUE [gr/m²] and SD of CPUE [gr/m²] values for three strata of Shirokë sub-basin

	0-3		3-6		> 6m	
	Average CPUE [gr/m2]	SD	Average CPUE [gr/m2]	SD	Average CPUE [gr/m2]	SD
Alburnus scoranza	4.269	2.002	8.419	2.342	6.474	2.969
Rutilus prespensis	3.074	3.656	4.155	3.266	4.091	3.524
Perca fluviatilis	0.255	0.622	0.254	0.565	0.576	1.401
Scardinus knezevici	0.306	0.753	0.055	0.071	0.013	0.020
Pseudorasbora parva	0.255	0.220	1.147	1.060	1.006	0.819
Carassius gibelio	0.381	0.641	0.339	0.893	0.059	0.205
Pachychilon pictum	0.636	0.564	1.112	0.898	0.772	0.725
Chondrostoma nasus	0.023	0.036	0.098	0.136	0.145	0.156
Cyprinus carpio	0.728	1.374	2.810	4.587	0.000	0.000
Rhodeus amarus	0.065	0.060	0.088	0.066	0.112	0.124
Allosa fallax	0.000	0.000	0.000	0.000	0.000	0.000
Cobitis meridionalis	0.003	0.007	0.000	0.000	0.000	0.000
Alburnoides ohridanus	0.716	1.202	0.027	0.060	0.000	0.000

As it is shown in Table 14, the biomass in Shiroka is more abundant in the 3-6 m depth compared to 0-3 and >6 m strata.

In the following graphs and table we are presenting CPUE in terms of numbers of individuals per one m² of MMG net. Those graphs are reflecting relative abundance of sampled species in Koplik sub-basin.



	0-3	3-6	> 6
Alburnus scoranza	0.340	0.489	0.389
Rutilus prespensis	0.342	0.386	0.354
Perca fluviatilis	0.003	0.008	0.009
Scardinus knezevici	0.015	0.040	0.009
Pseudorasbora parva	0.067	0.356	0.306
Carassius gibelio	0.010	0.004	0.002
Pachychilon pictum	0.094	0.121	0.074
Chondrostoma nasus	0.014	0.030	0.043
Cyprinus carpio	0.010	0.024	0.000
Rhodeus amarus	0.048	0.053	0.050
Allosa fallax	0.000	0.000	0.000
Cobitis meridionalis	0.003	0.000	0.000
Alburnoides ohridanus	0.015	0.012	0.000

Figure 60. CPUE ratio and absolute average CPUE [ind/m²] per three strata of Shirokë sub-basin

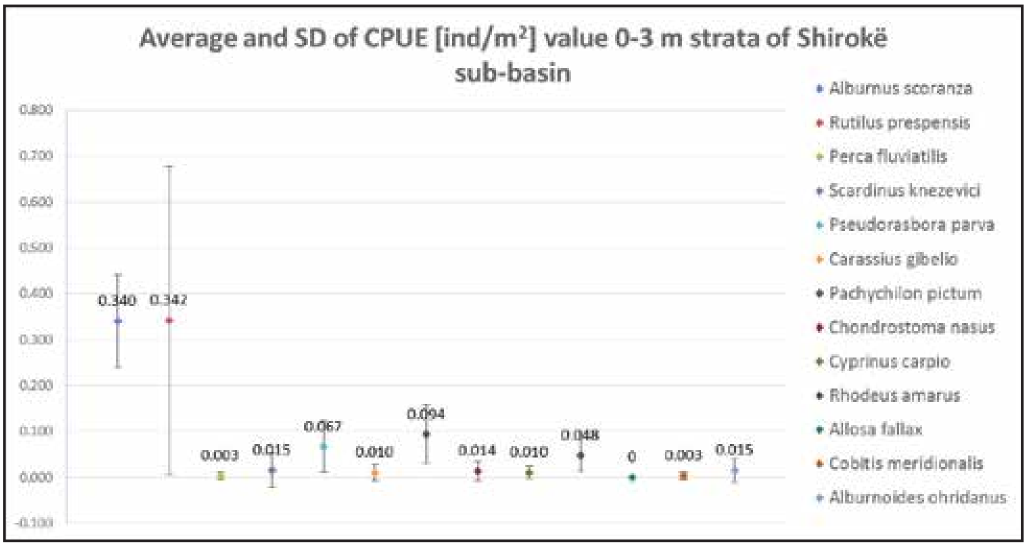


Figure 61. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value 0-3m strata of Shirokë sub-basin

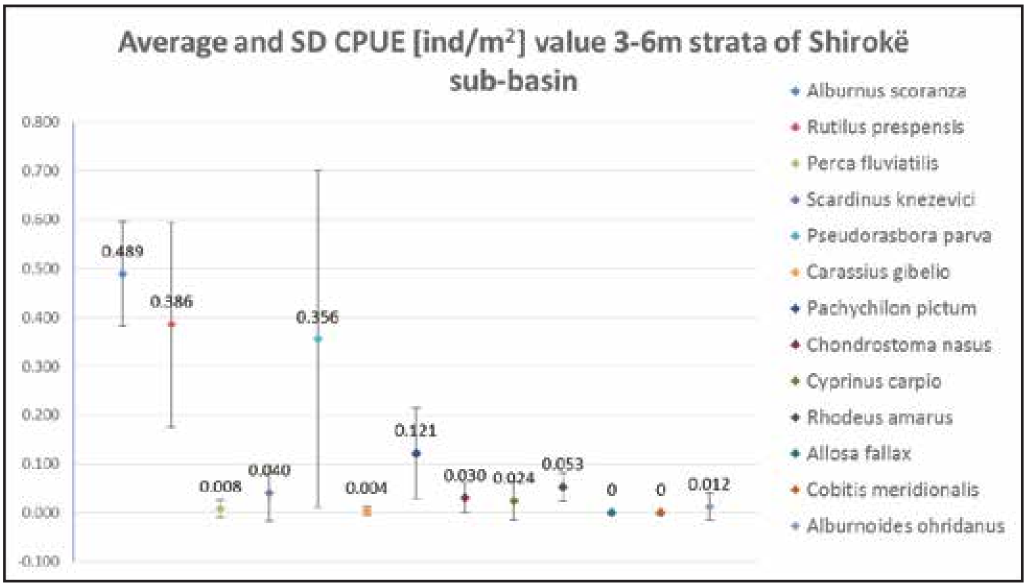


Figure 62. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value 3-6m strata of Shirokë sub-basin

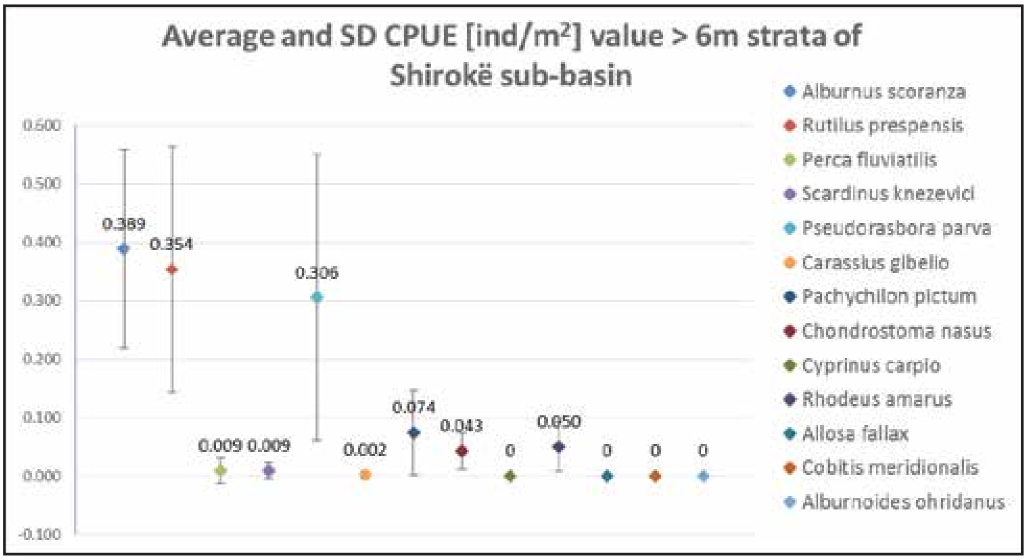


Figure 63. Average CPUE [ind/m²] and SD of CPUE [ind/m²] value >6m strata of Shirokë sub-basin

Table 15. Average CPUE [ind/m²] and SD of CPUE [ind/m²] values for three strata of Shirokë sub-basin

	0-3		3-6		> 6m	
	Average CPUE [ind/m2]	SD	Average CPUE [ind/m2]	SD	Average CPUE [ind/m2]	SD
Alburnus scoranza	0.340	0.101	0.489	0.107	0.389	0.170
Rutilus prespensis	0.342	0.335	0.386	0.210	0.354	0.210
Perca fluviatilis	0.003	0.008	0.008	0.018	0.009	0.022
Scardinus knezevici	0.015	0.038	0.040	0.058	0.009	0.015
Pseudorasbora parva	0.067	0.056	0.356	0.345	0.306	0.244
Carassius gibelio	0.010	0.017	0.004	0.009	0.002	0.006
Pachychilon pictum	0.094	0.064	0.121	0.092	0.074	0.072
Chondrostoma nasus	0.014	0.021	0.030	0.030	0.043	0.031
Cyprinus carpio	0.010	0.015	0.024	0.040	0.000	0.000
Rhodeus amarus	0.048	0.035	0.053	0.029	0.050	0.041
Allosa fallax	0.000	0.000	0.000	0.000	0.000	0.000
Cobitis meridionalis	0.003	0.008	0.000	0.000	0.000	0.000
Alburnoides ohridanus	0.015	0.026	0.012	0.027	0.000	0.000

If we consider Table 15, depth strata 3-6 m and > 6 m are similar in number while 0-3 m depth has lower values. This means that some large individuals (especially from C. carpio) were present in the lower depth and has contributed to the biomass values.

4.3.2.1.3. Population structure

Based on MMG gillnet data sets (0-3, 3-6 and >6m m strata) we can analyze populations of the five most abundant species that also has highest CPUE values. Other species individuals were not present in such numbers in sample and it was not possible to do following analysis for theme. We have used one cm length classes for each of the species we analysed.

Alburnus scoranza **(bleak)**

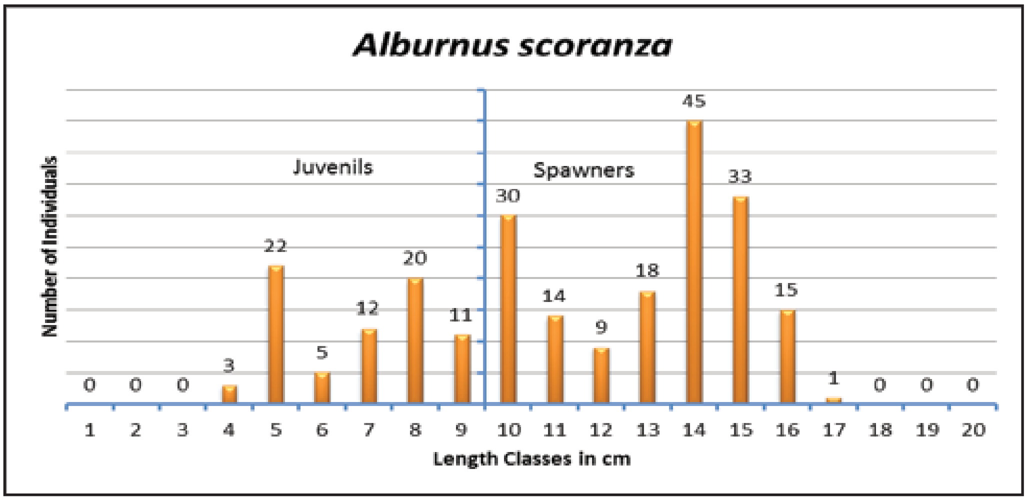


Figure 64. Length frequency of bleak (Alburnus scoranza) in total catch of Koplik sub-basin

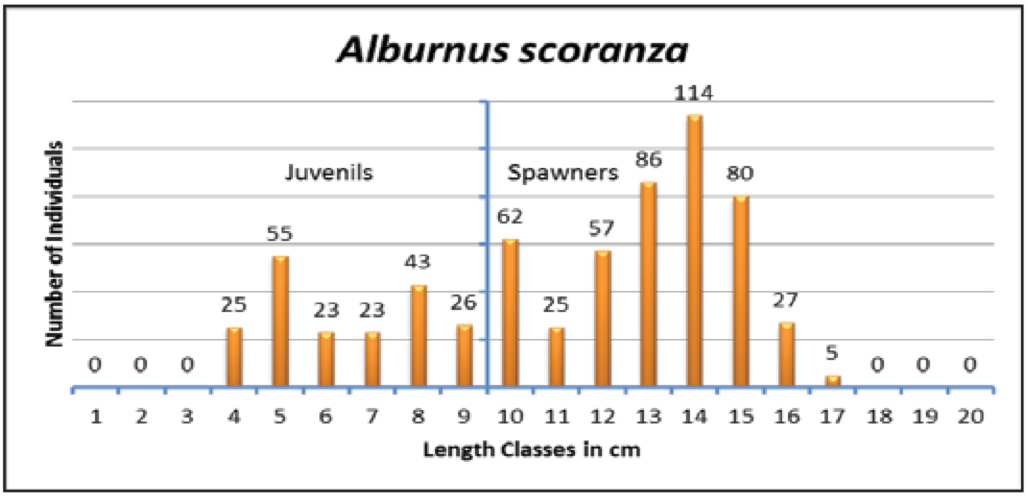


Figure 65. Length frequency of bleak (Alburnus scoranza) in total catch of Shiroke sub-basin

From previous figures it is clear that we, in our sample, have had all age cohorts of bleak with dominance of fish with length in intervals of 13-16cm and 8-11cm respectively. Such length frequency composition completely correspondent with bleak abundance considering that in both areas this species appears more abundant in deeper strata where older fish individuals occur. Also there is not big differences in the age distribution in both areas, except for the fact that Shiroka has bigger numbers in terms of catch.

Perca fluviatilis **(perch)**

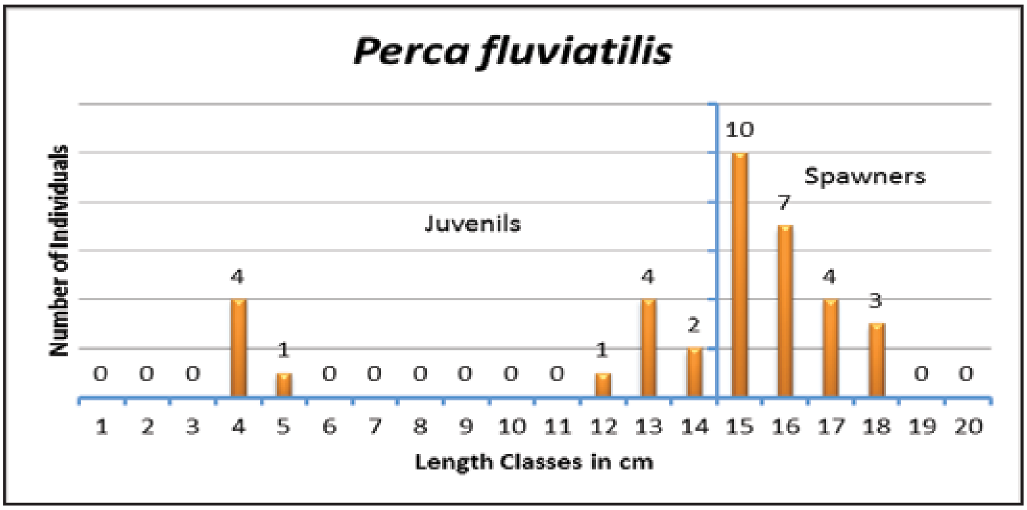


Figure 66. Length frequency of perch (Perca fluviatilis) in total catch of Koplik sub-basin

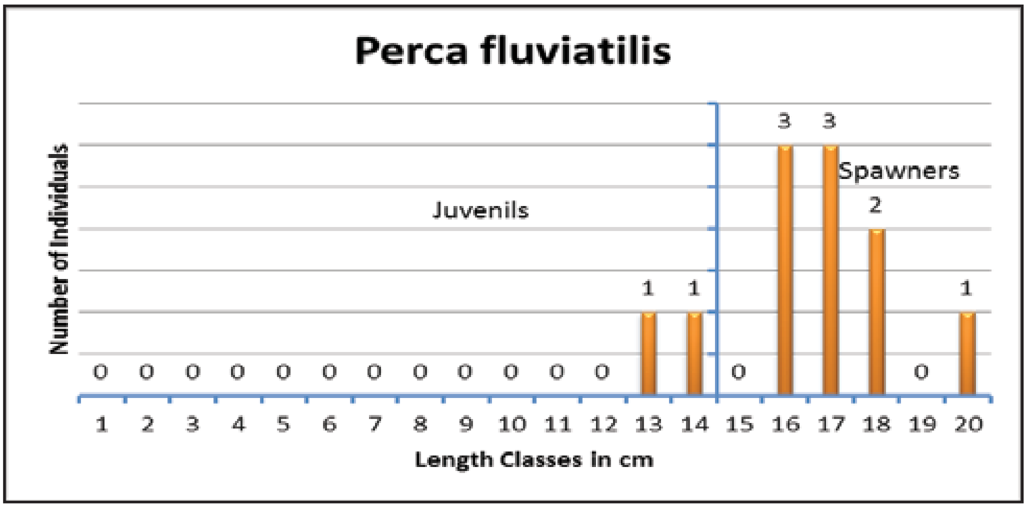


Figure 67. Length frequency of perch (Perca fluviatilis) in total catch of Shiroke sub-basin

From previous figures, it is clear that we have had the dominance of middle size fish in our sample (length class interval 15-18 cm) indicating of dominance 1^+ and 2^+ age cohorts.

Rutilus prespensis **(roach)**

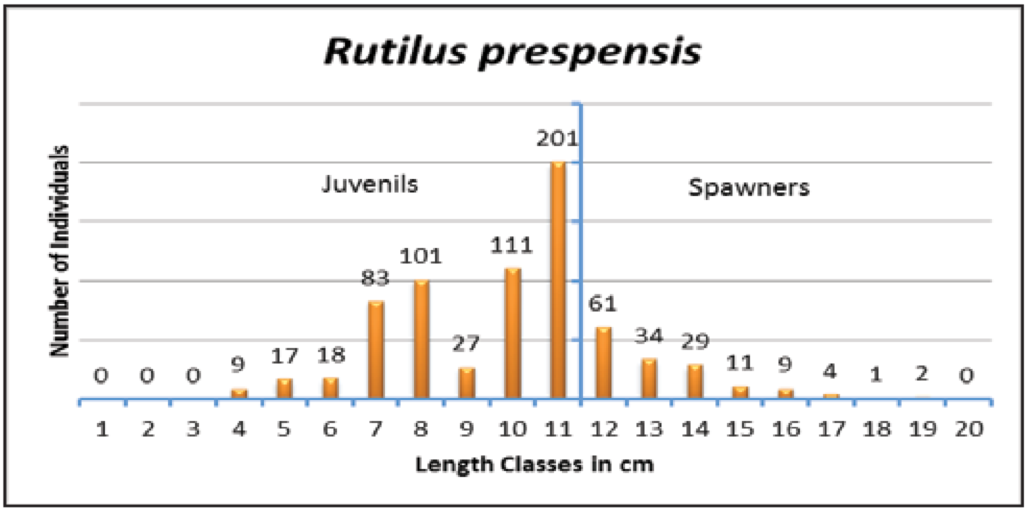


Figure 68. Length frequency of roach (R. prespensis) in total catch of Koplik sub-basin

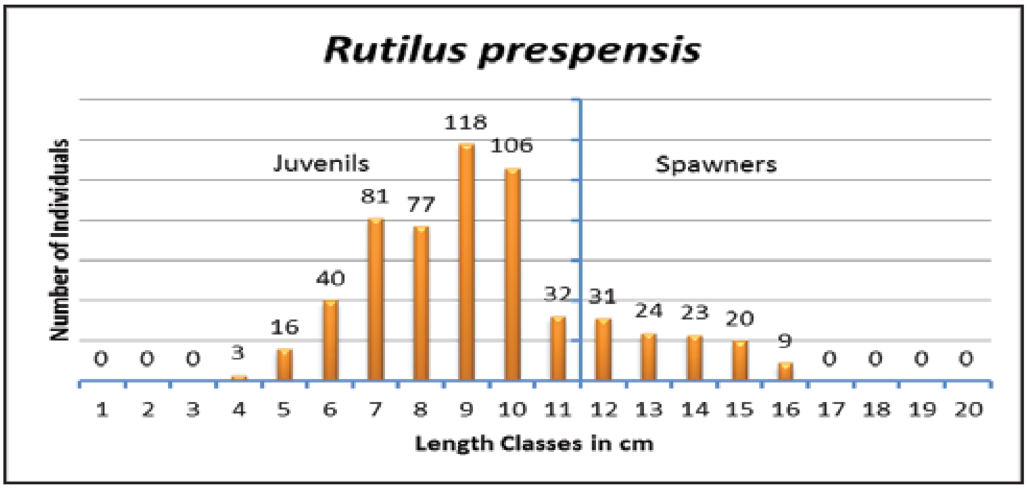


Figure 69. Length frequency of roach (R. prespensis) in total catch of Shiroke sub-basin

From previous graph it is clear that in our sample dominant were individuals from 7-10 and 10-13 cm of length intervals suggesting dominance of 1^+ and 2^+ age cohorts that indicate good population structure. No significant differences were noted in the population structure between Shiroka and Koplik, but no large size individuals (+16 cm) were observed in both areas, while in MNE side these individuals were frequent.

Pachychilon pictum

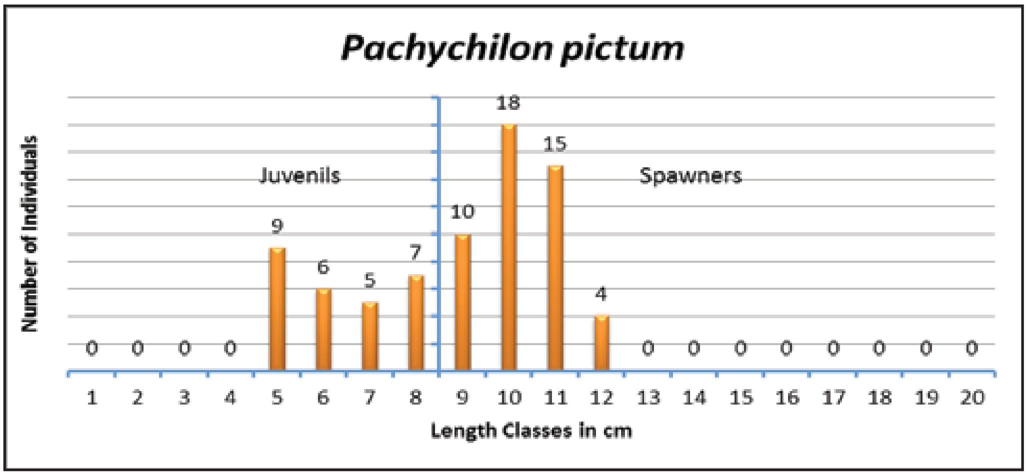


Figure 69. Length frequency of Pachychilon pictum in total catch of Koplik sub-basin

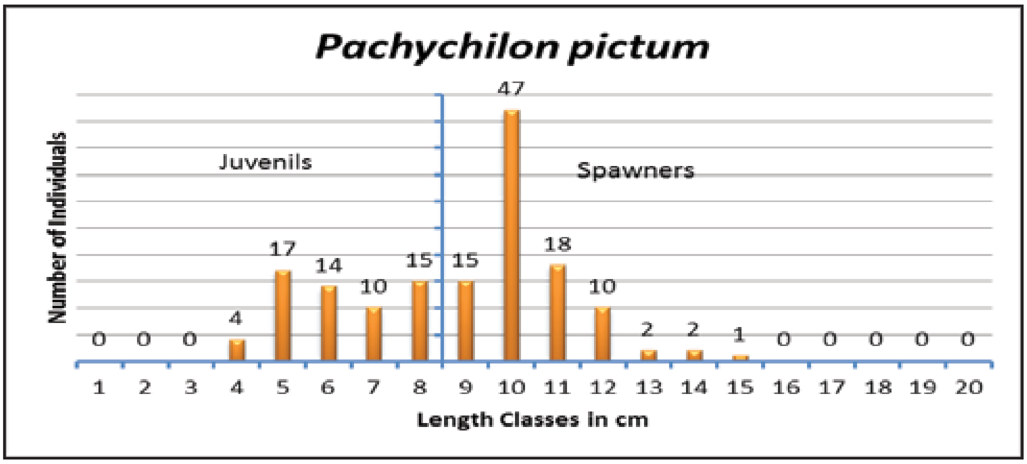


Figure 70. Length frequency of Pachychilon pictum in total catch of Shiroke sub-basin

From previous figure it is clear that we have had dominance of fish from 5-7cm, 8-10cm and 10-12cm length intervals respectively suggesting good population structure since youngest age cohort ascendency.

Pseudorasbora parva

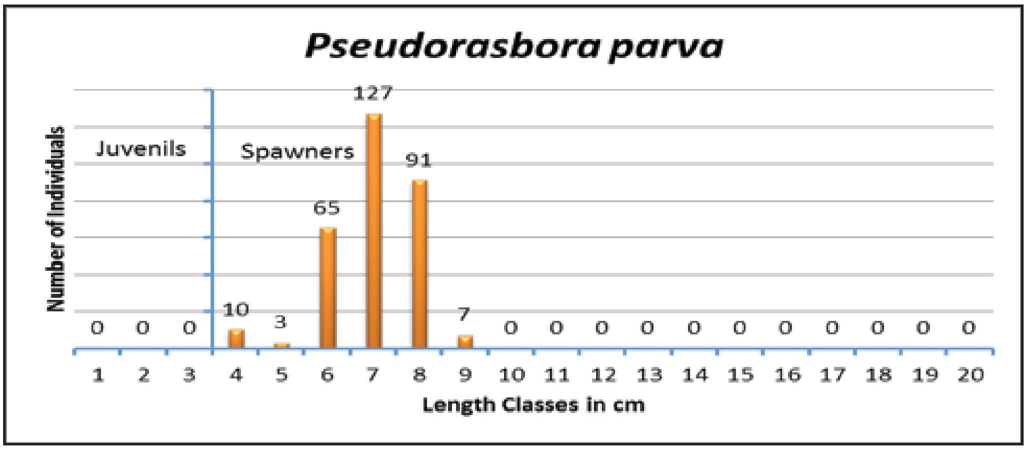


Figure 71. Length frequency of Pseudorasbora parva in total catch of Koplik sub-basin

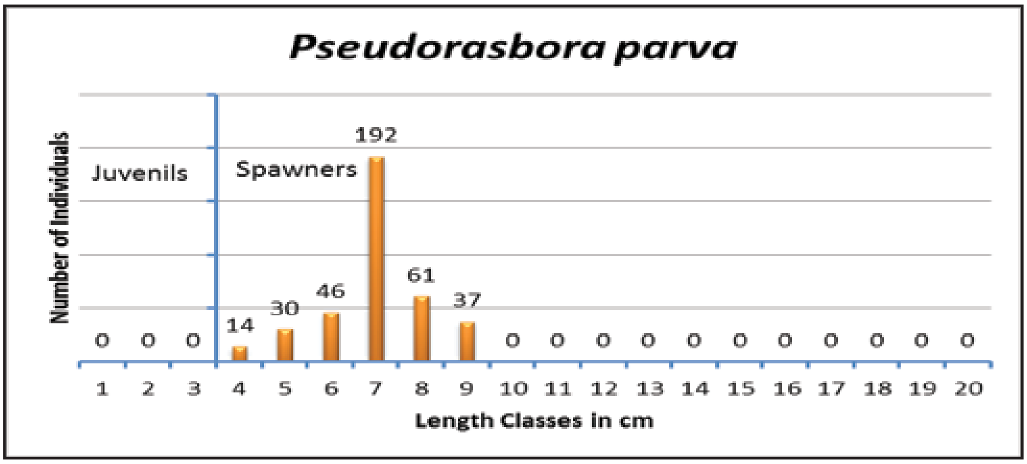


Figure 72. Length frequency of Pseudorasbora parva in total catch of Shiroke sub-basin

There is a total dominance of spawners of P. parva both in Koplik and Shiroka but this is attributable to the small size of juveniles of these species that rarely enter in the MMG net catchability.

Rhodeus amarus

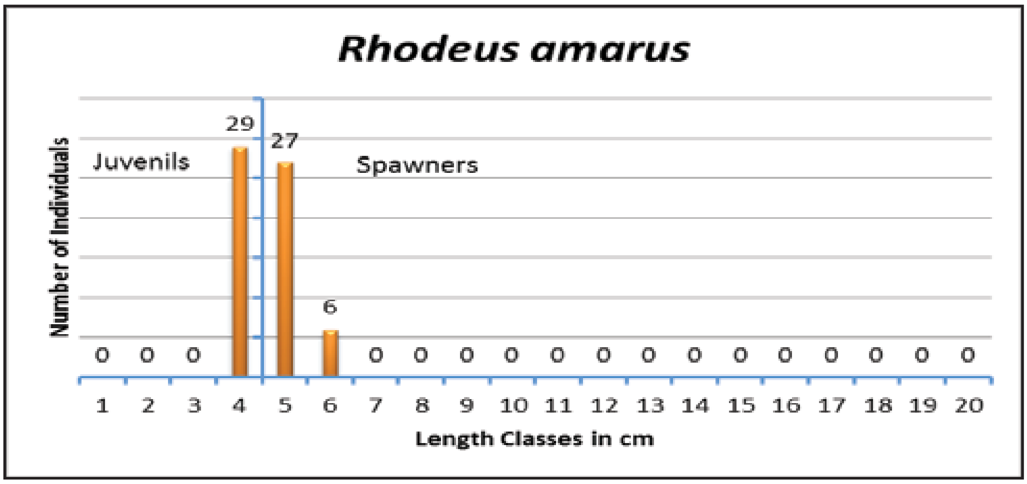


Figure 73. Length frequency of Rhodeus amarus in total catch of Koplik sub-basin

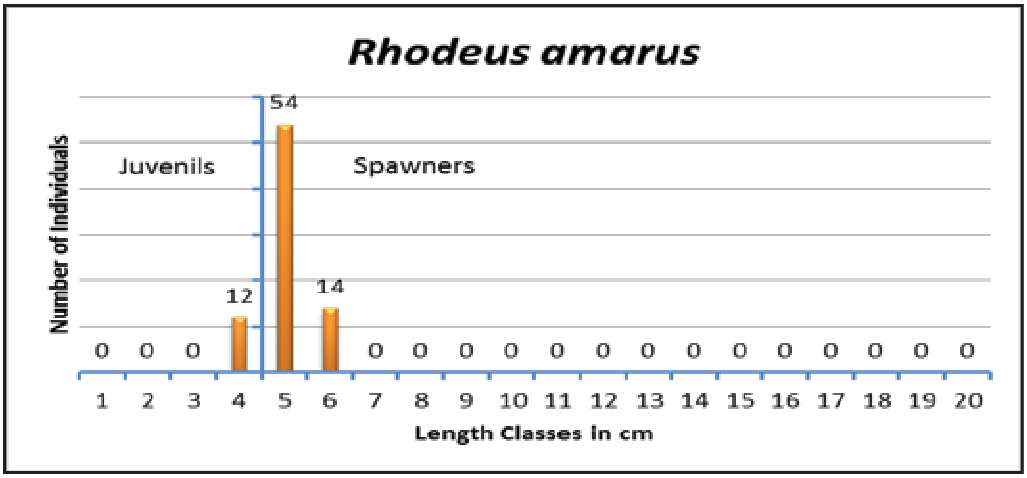


Figure 74. Length frequency of Rhodeus amarus in total catch of Shiroke sub-basin

There is a dominance of spawners of R. amarus both in Koplik and Shiroka but this is attributable to the small size of juveniles of these species that rarely enter in the MMG net catchability.

5. Discussion

5.1. MMG nets

The results of this study strongly support conclusion that Skadar/Shkodra Lake fish fauna differs in terms his northeastern (Vranjina-Virpazar-Grmožur sub-basin) and southwestern (Shiroka and Koplik sub-basins) parts. Differences in detected fish fauna are most obvious in 0-3m strata while they are less dramatic in deeper 3-6m stratum (Figures 75 and 76)

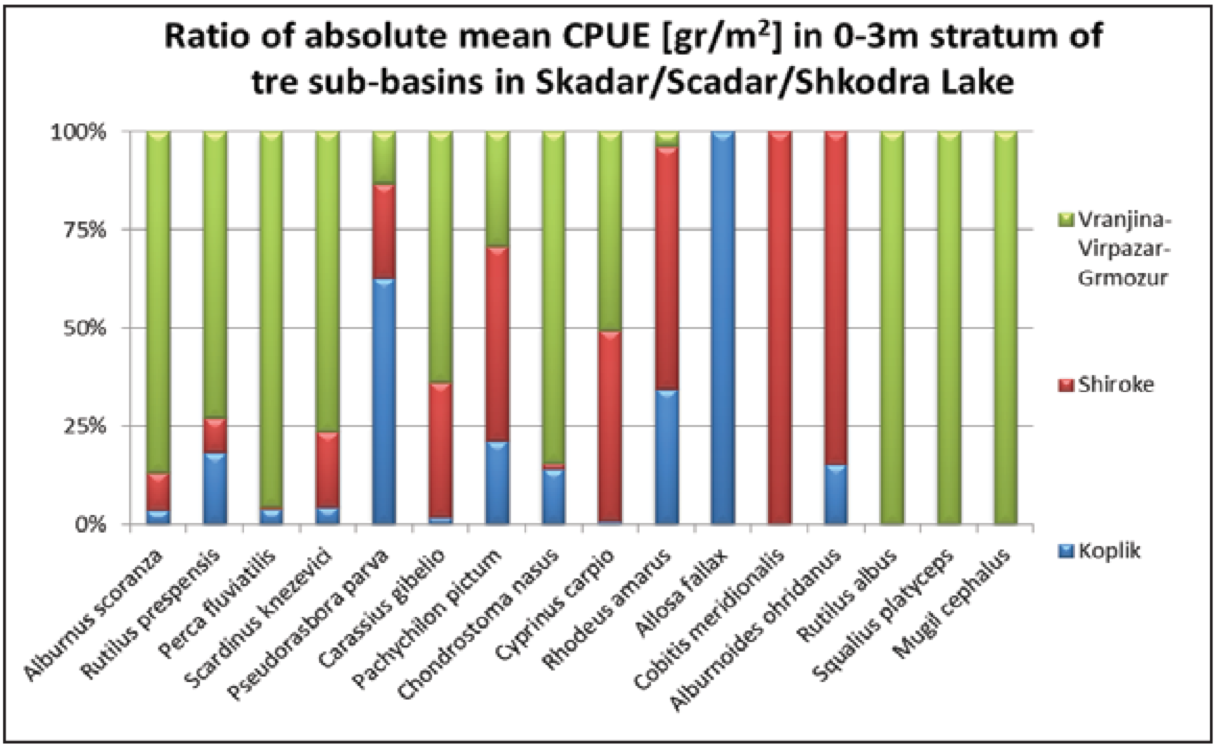


Figure 75. Individual species CPUE [gr/m^2] ratio in 0-3m stratum among all three sub-basins researched in Skadar/Shkodra (MMG nets)

Those differences are dominantly reflecting in detected relative biomass of sampled species. Considering 0-3m stratum, only 3 of 10 common species shows higher biomass in Albanian part (Pseudorasbora parva, Pachychilon pictum and Rhodeus amarus), common carp shows equal biomass but it was absent in Koplik sub-basin while bleak, roach, perch, Scardinius knezevici, silver carp and nase had unexpectedly higher CPUE values in MNE sub-basin. Moreover differences in this stratum are in species structure also. In Albanian side there were three species which were absent in Montenegrin side (Alosa falax, Cobitis meridionalis and Alburnoides ohridanus) while the opposite situation was with Rutilus albus, Squalius platyceps and Mugil cephalus. All in all in 0-3m stratum in Skadar/Shkodra Lake, with MMG nets, we sampled 16 fish species.

Table 16. Individual species absolute CPUE [gr/m^2] in 0-3m stratum for all three sub-basins researched in Skadar/Shkodra (MMG nets)

	Koplik	Shiroke	Vranjina-Virpazar-Grmozur
Alburnus scoranza	1.631	4.269	37.998
Rutilus prespensis	6.281	3.074	25.128
Perca fluviatilis	1.690	0.255	39.488
Scardinus knezevici	0.073	0.306	1.219
Pseudorasbora parva	0.662	0.255	0.141
Carassius gibelio	0.020	0.381	0.704
Pachychilon pictum	0.272	0.636	0.373
Chondrostoma nasus	0.204	0.023	1.204
Cyprinus carpio	0.016	0.728	0.762
Rhodeus amarus	0.036	0.065	0.004
Allosa fallax	0.177	0.000	0.000
Cobitis meridionalis	0.000	0.003	0.000
Alburnoides ohridanus	0.131	0.716	0.000
Rutilus albus	0.000	0.000	0.934
Squalius platyceps	0.000	0.000	1.141
Mugil cephalus	0.000	0.000	0.169

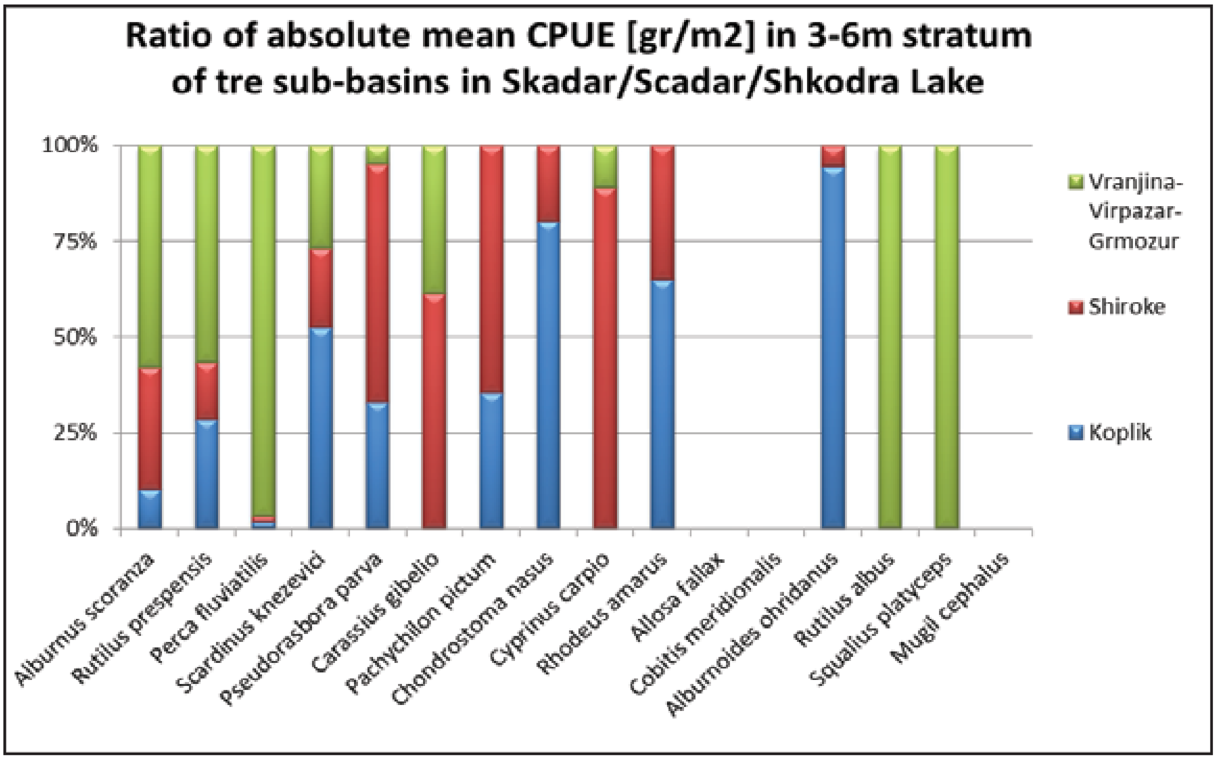


Figure 76. Individual species CPUE [gr/m^2] ratio in 3-6m stratum among all three sub-basins researched in Skadar/Shkodra Lake (MMG nets)

In 3-6m stratum situation is slightly different but still showing great divergence. Like in shallower part, bleak, roach and perch showed unexpectedly higher CPUE values in MNE side while Pseudorasbora parva and common carp shows just opposite situation (greatly higher CPUE in AL side, in Koplik common carp was absent). Pachychilon pictum was absent in deeper stratum in MNE as the nase and Rhodeus amarus. In AL side Rutilus albus and Squalius cephalus were absent like in shallow stratum while the same situation was with Alburnoides ohridanus on MNE side. Comparing with 0-3 stratum Alosa fallax, Cobitis meridionalis and Mugil cephalus was absent so we detected 13 fish species in this 3-6m Skadar/Shkodra Lake layer.

Table 16. Individual species absolute CPUE [gr/m²] in 3-6m stratum for all three sub-basins researched in Skadar/Shkodra Lake (MMG nets)

	Koplik	Shiroke	Vranjina-Virpazar-Grmozur
Alburnus scoranza	2.647	8.419	15.146
Rutilus prespensis	7.833	4.155	15.600
Perca fluviatilis	0.261	0.254	15.995
Scardinus knezevici	0.139	0.055	0.071
Pseudorasbora parva	0.609	1.147	0.091
Carassius gibelio	0.000	0.339	0.213
Pachychilon pictum	0.611	1.112	0.000
Chondrostoma nasus	0.392	0.098	0.000
Cyprinus carpio	0.000	2.810	0.346
Rhodeus amarus	0.162	0.088	0.000
Allosa fallax	0.000	0.000	0.000
Cobitis meridionalis	0.000	0.000	0.000
Alburnoides ohridanus	0.470	0.027	0.000
Rutilus albus	0.000	0.000	0.203
Squalius platyceps	0.000	0.000	1.264
Mugil cephalus	0.000	0.000	0.000

Considering relative abundance CPUE [ind/m²] we detected similar situation as for relative biomass values (Figure 77 and 78). The same species were fare more abundant in MNE part of 0-3m stratum likes for biomass values (bleak, perch, Scardinius knezevici and roach). In MNE part nase and common carp shows less values comparing to same stratum in AL (Koplik and Shiroka) suggesting presence of bigger individuals in MNE part while for silver carp we had just opposite situation.

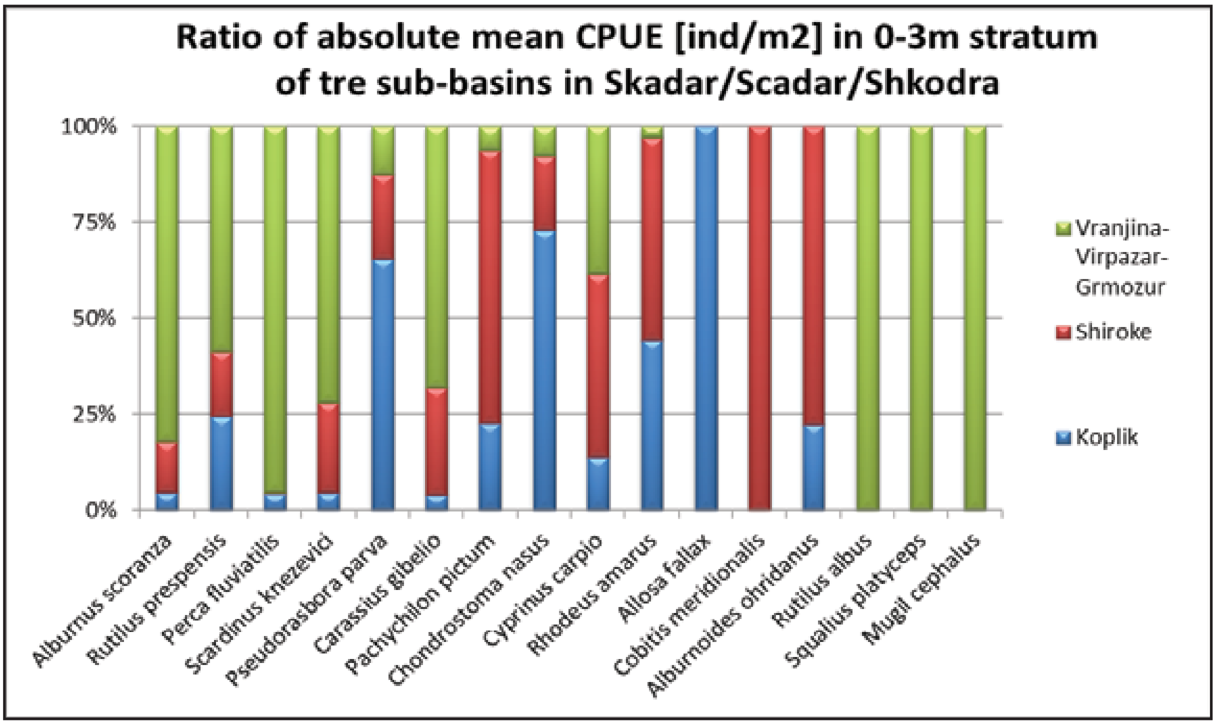


Figure 77. Individual species mean CPUE [ind/m²] ratio in 0-3m stratum among all three sub-basins researched in Skadar/ Shkodra Lake (MMG nets)

Table 17. Individual species absolute CPUE [ind/m^2] in 0-3m stratum for all three sub-basins researched in Skadar/Shkodra Lake (MMG nets)

	Koplik	Shirokë	Vranjina-Virpazar-Grmozur
Alburnus scoranza	0.119	0.340	2.117
Rutilus prespensis	0.486	0.342	1.172
Perca fluviatilis	0.039	0.003	0.872
Scardinus knezevici	0.003	0.015	0.047
Pseudorasbora parva	0.200	0.067	0.039
Carassius gibelio	0.001	0.010	0.025
Pachychilon pictum	0.030	0.094	0.008
Chondrostoma nasus	0.052	0.014	0.006
Cyprinus carpio	0.003	0.010	0.008
Rhodeus amarus	0.040	0.048	0.003
Allosa fallax	0.050	0.000	0.000
Cobitis meridionalis	0.000	0.003	0.000
Alburnoides ohridanus	0.004	0.015	0.000
Rutilus albus	0.000	0.000	0.025
Squalius platyceps	0.000	0.000	0.006
Mugil cephalus	0.000	0.000	0.003

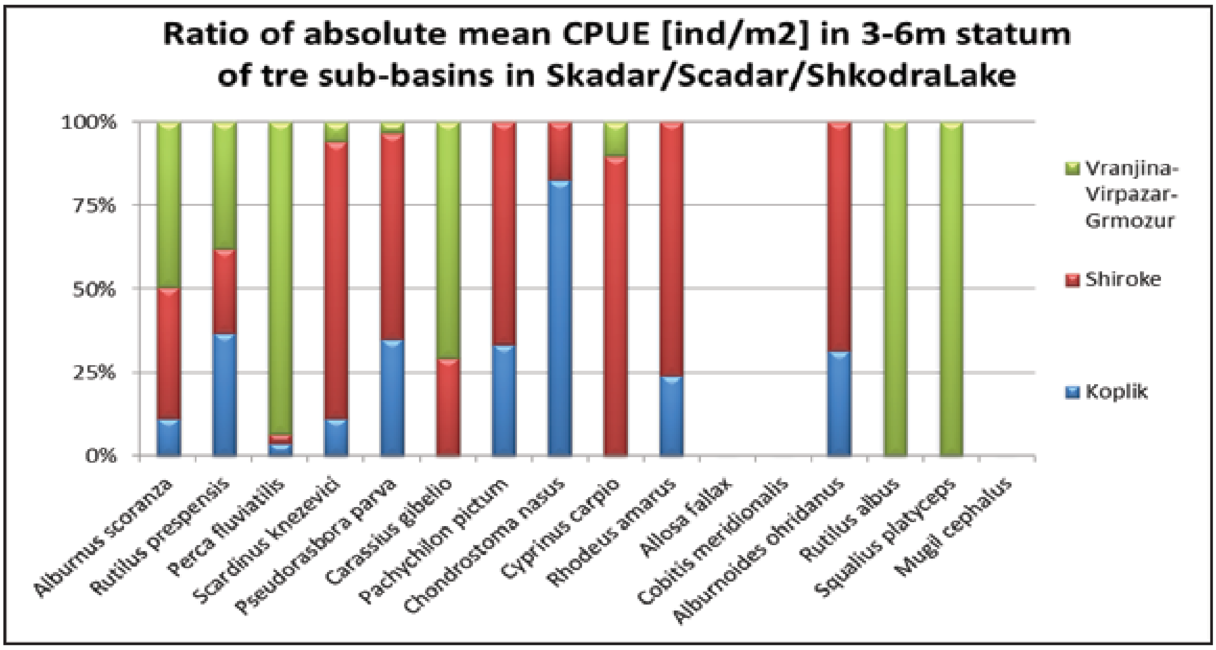


Figure 78. Individual species CPUE [ind/m^2] ratio in 3-6m stratum among all three sub-basins researched in Skadar/Shkodra Lake(MMG nets)

If we compare abundance and biomass values for 3-6m stratum of we had the similar tendency. In MNE sub-basin, in contrast to biomass, abundance values for Scardinius knezevići and bleak and roach are smaller sub-basin suggesting slightly bigger fish individuals. Situation with silver carp is opposite suggesting slightly bigger individuals in AL sub-basins of the same stratum.

Table 18. Individual species absolute mean CPUE [ind/m²] in 3-6m stratum for all three sub-basins researched in Skadar/Shkodra Lake (MMG nets)

	Koplik	Shirokë	Vranjina-Virpazar-Grmozur
Alburnus scoranza	0.143	0.489	0.621
Rutilus prespensis	0.559	0.386	0.575
Perca fluviatilis	0.011	0.008	0.274
Scardinus knezevici	0.006	0.040	0.003
Pseudorasbora parva	0.202	0.356	0.019
Carassius gibelio	0.000	0.004	0.010
Pachychilon pictum	0.061	0.121	0.000
Chondrostoma nasus	0.144	0.030	0.000
Cyprinus carpio	0.000	0.024	0.003
Rhodeus amarus	0.017	0.053	0.000
Allosa fallax	0.000	0.000	0.000
Cobitis meridionalis	0.000	0.000	0.000
Alburnoides ohridanus	0.006	0.012	0.000
Rutilus albus	0.000	0.000	0.007
Squalius platyceps	0.000	0.000	0.004
Mugil cephalus	0.000	0.000	0.000

The reasons for such differences in fish fauna composition (species structure, biomass and abundance of detected species) we can find in two possibilities. First one is ecological differences in two Lake parts. MNE part is under strong influence of fresh water from one big tributary (Morača river), several smaller (Rijeka Crnojevica, Orahovštica, Karatuna and Crminčka river) and numerous external and sub-lacustrine springs, which is not the situation with AL part (especially in south-eastern part of the Lake). The second reason could be overfishing which is spatially related to favorable fishery species such are bleak, nase, chub, common and silver carp while decreasing of other specie could be explained as consequences of by-catch during heavy fishery activity.

Different composition of sampled specie could be explained with differences in ecological parameters also. Namely, presence of Alosa falax only in AL sub-basins is related to vicinity of Buna/Bojana River which connects Lake with Adriatic Sea. But here we have to stress that absence of same species in MNE sub-basin could be explained with lower population number due to less favorable habitats and fishery pressure since we are convinced in the presence of this species in MNE part. Presence of Cobitis meridionalis only in Shiroka sub-basin could indicate slight eutrophication in this part of the Lake where this fish showed higher population density due to favorable ecological conditions. We are sure that this fish occurs in other two sub-basins but with low population density so we haven't it in our MMG nets. The presence of Rutilus albus only in MNE part is consequence of this species ecology since white roach prefer Lake water-mass under stronger influence of springs and smaller tributaries. In addition to this reason we can also add the fact this species is described relatively lately and that it is highly similar to common Lake roach what make his determination relatively difficult. Absence of Alburnoides ohridanus in MNE sub-basin is consequence low abundance in this Lake part but also in less favorable ecological conditions since in some past research we had this fish in our catch on MNE side. Absence of chub and gray mullet in AL sub-basins could be explained only with their low abundance due to overfishing. It was logical that grey mullet, as twaite-shad, occurs in higher numbers in Lake part closer to marine ecosystem than in those that are distance to Adriatic sea but according to our research it seems very rare in this region since there was no individuals in MMG catch of AL sub-basins. Similar was with chub since this fish species is almost so called "cosmopolite" species that lives in both, fresh water riverine but also in all Lake habitats.

Considering WFD and fish as bio-indicators of water body quality (status), from this research we can assume that north-western parts of the Lake has a lower level of eutrophication comparing to south-eastern Lake parts. Since an historical data on fish assemblages of the Lake are mostly related to fishery sector we can assume that our findings correlate with theme in terms of abundance and biomass of most common species in Lake. Low CPUE values for common and silver carp are consequence of MMG nets selectivity, those two fish growing and behavior patterns as well as their body shape causing low netting possibility (in standard MMG nets biggest mesh size is 55 mm). Speaking in general for whole Lake we can say that our results in first year of standardized monitoring with MMG nets shows expected findings since we hypothesize that structure of Lake fauna haven't been changed a lot in terms of their structure but we found unexpected changes in terms of quality (biomass and abundance). Future monitoring have to give precise answer weather this was some normal natural circle or it was trend that we detected. Our standing point is that final answer and more precise characterization toward fulfilling of WFD could be given only after at least 5 years of the same standardized MMG monitoring.

5.2. Electro-fishing transects

The purpose of using this methodology was our intention to sample bigger fish size species, especially those of older age cohort that was impossible to catch with standard MMG nets. This methodology was applied only on MNE side as a supplementary to standard MMG sampling. Due to project starting delay of several months (because of problems in different gear procurement) we haven't be able to apply additional electro-fishing transect in February so results are those from September campaign.

Research shows that among so called big body sized species, common carp is still most dominant in terms of biomass while chub is most abundant (Figures 79 and 80). This is something that is in coherence with data from the past that was related to fishery total catch on yearly level. This indicates that Lake is still relatively stable in terms of fish fauna composition. Major differences comparing with old fishery data from the past century are in rising of biomass and abundance of alien silver carp species and domestic chub while on the other side we detected decreasing of biomass and abundance of nase and gray mullet. In last several years in our research we haven't encounter mullet and nase in our samples, and this research is something that encourages us to state that this year sampling of those two species point in the direction of a kind of Lake recovery.

Our opinion is that final answer and more precise characterization toward fulfilling of WFD could be given only after at least 5 years of the same standardized electro-fishing transect monitoring. Additionally, we think that this methodology should be applied in AL side also and that should serve as additional methodology for fulfilling of WFD demands.

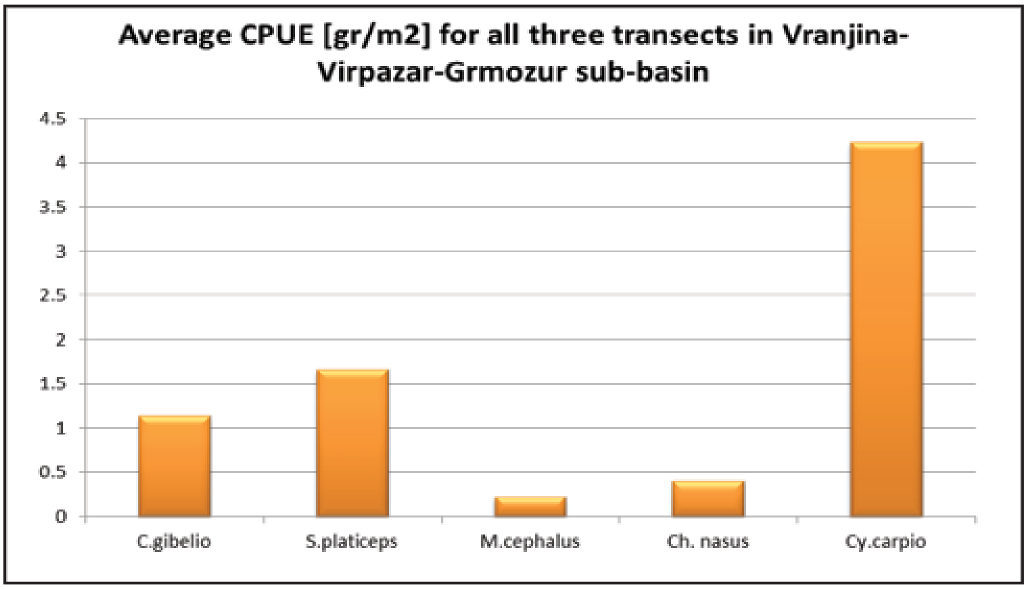


Figure 79. Average relative biomass CPUE [gr/m2] for all three electrofishing transects in Vranjina-Virpazar-Grmozur sub-basin

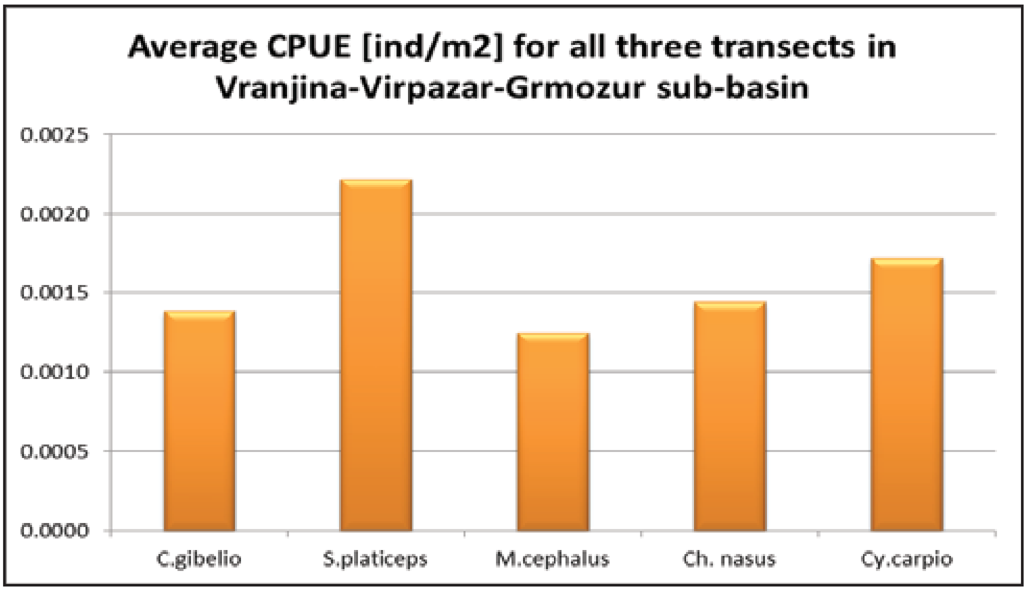


Figure 80. Average relative abundance CPUE [ind/m2] for all three electrofishing transects in Vranjina-Virpazar-Grmozur sub-basin

Table 18. Absolute average relative biomass CPUE [gr/m²] and absolute average relative abundance CPUE [ind/m²] for five big body size fish species in Vranjina-Virapazar-Grmožur sub-basin.

	C.gibelio	S.platiceps	M.cephalus	Ch. nasus	Cy.carpio
Average CPUE[gr/m²]	1.1325	1.6582	0.2221	0.4019	4.2380
Average CPUE [ind/m²]	0.0014	0.0022	0.0013	0.0014	0.0017

5.3. Commercial bleak harvesting

Monitoring of commercial bleak harvesting showed that in 2013 bleak represent 83% of kalimera catch while in 2014 share of bleak was 68 %. In 2013 as by-catch there were present additional three species (roach, perch and chub) while in 2014 there were five of theme: roach, perch, silver carp, common carp and trout (Figures 36 and 37).

In terms of fishery we found out that commercial catch is composed of almost higher cohort individuals (spawners, 99,9 %) which excellent from sustainable resource usage point of view. We detected that mesh-size of so called kalimera net was not 15.5 mm measured “knot to knot” (it was smaller, about 14 mm) regarding stretched diagonal measurement so the application of this rule will allow long term usage of bleak resource from the Lake.

Sampling of trout in lake “oka” suggests good ecological status of the Lake from WFD point of view. In terms of fishery monitoring as well as in frame of WFD needs continuous monitoring of commercial harvesting in future is of great importance. Since in AL side there is no stationary commercial harvesting of bleak monitoring of beach seining would be helpful also. Our opinion is that purchasing of beach seine net would be of great importance since researcher could do experimental seining and we will avoid any problems with fisherman and concessioners since they have to announce such fishing to management authorities. Additionally we had information that seining most commonly happened illegally over the night (at least in MNE part) and beach seine net will allow us, under supervision of Lake Ranger services, to implement this kind of sampling without any obstacles. Moreover we think that monitoring of catch from V shaped fish weir tool on Bojana/Buna river should be also conveyed.

Considering monitoring of fish larvae in Lake we hope that in following years using of experimental beach seine for larvae during summer months will allow us to motorize this important aspect on both sides.

6. Conclusions

Application of MMG gillnets and EN 14757 standards allow, for the first time after almost 40 years, systematic approach in investigation of Skadar/Shkodra Lake fish fauna and fisheries. They show excellent potential when comparison with other Balkan lakes (but not only Balkan) as well as the comparison of long time monitoring serials is about.

Bleak (Alburnus scoranza) is most abundant species in terms of numbers of individuals as well as in terms of biomass, in both Skadar/Shkodra Lake strata in MNE part. Perch (Perca fluviatillis) turned out to be second by biomass in each strata of the Skadar/Shkodra Lake (CPUE) of all sampled species in Vranjina-Virpazar-Grmožur sub-basin. In Shiroka sub-basin (AL part of the Lake) bleak (A. scoranza) was dominant in terms of biomass and abundance while in Koplik sub-basin (AL part of the Lake) roach (Rutilus prespensis) shows highest biomass and abundance. In deeper strata we detect less abundance but individuals with larger body size (older age group members) of bleak, roach and perch in all sub-basins from Skadar/Shkodra Lake.

Absolute CPUE values for bleak, roach and perch was unexpectedly higher on MNE side of the lake suggesting higher abundance and biomass. Such situation could be caused by ecological differences in two Lake parts. MNE part is under strong influence of fresh water from one big tributary (Morača river), several smaller (Rijeka Crnojevica, Orahovštica, Karatuna and Crminčka river) and numerous external and sub-lacustrine springs, which is not the situation with AL part (especially in south-eastern part of the Lake). Additional reason could be overfishing which is spatially related to favorable fishery species such are bleak, nase, chub, common and silver carp while decreasing of other specie could be explained as consequences of by-catch during extensive fishery.

MMG gill nets shows low capacity for netting of species with faster growth and with bigger body size (common carp, chub, nase, mullet, silver carp) and electro-fishing gives more detail insight in population structure of those species. Population structure of the most abundant species in MMG samples shows normal distribution of age cohorts and no signs of recent overfishing events.

Carp samples from electro-fishing transect shows relatively good abundance and biomass considering high fishery pressure because this species is one of the two most important fish species in fishery sense. Legally proscribed lowest mesh size for bleak in MNE (15,5 mm) shows as a good choice since that in 15,5 mm panels we had only individuals larger than 12 cm of TL. Population structure from the harvesting subsample (MNE part of the lake) shows certain selectivity of "kalimera" net regarding absence of juvenile age class of bleak. Standardized "knot to knot" measuring (not diagonal) as well as the application of appropriate legislation (15,5mm mesh size) will restrict catch of bleak on individuals that are not smaller than 12 cm. This will allow sustainable and long term usage of bleak resource. Proscribed minimal allowed length for bleak in MNE is completely wrong regarding that lowest allowed length for bleak is 16cm. According to our data this length class is almost absent and biggest biomass of bleak is between 12 and 15 cm length. Beach seining of bleak ("gribljenje") is completely uncontrolled and unregulated activity in MNE part of Skadar/Shkodra Lake and source of poaching of huge fish mass. Moreover, seining also take out from the lake water a big amount of small carp (0^+ and 1^+ age cohorts) every year. Panels of MMG nets with mesh size 19,5 -24 mm exclusively catch only bigger perch and bigger roach and shows not only selectivity for TL but also selectivity for species making such net favorable fishing gear even during fishing ban period.

Application of this monitoring scheme and methodology provide us a good basis regarding WFD needs in terms of determination of fish population. Diversification of sampling localities, gears in use and sampling period will allow more diverse sample in terms of fish species, especially those that are of importance for WFD. We suggest to implement electro-fishing transect on AL side as well as experimental larvae and beach seining on both side and monitoring of catch from V shape fish wire traps on Bojana/Buna river. Since we failed in sampling of eel our suggestion is to use so called "pari" gear or longline gear (long basic line or rope with 50-100 hooks laterally attach on it on each 1,5m and posed on Lake bottom).

Different composition of sampled species could be explained with differences in ecological parameters. Northwestern part of the lake is abundant in fresh water (one big inlet – Morača River, numerous middle and small size inlets and numerous external and sub-lacustrine springs and wells), characterized with divers Lake bottom substrate and whit vast flooding area along whole northern coast. Southeastern part of the Lake is mainly characterized with less impact of the fresh water, Lake bottom is mainly made of mud and stone, smaller flooding area and relatively closeness of marine habitats trough Buna/Bojana river as outlet. Fish species structure suggest that northwestern part of the lake is more oligotrophic than southeastern part which is caused by less impact of fresh water but also by more dense human settlements along lake coast (city of Shkodra as the largest settlement).

Presence of trout in our samples suggests good ecological status of the Lake from WFD perspective. Major differences comparing with old fishery data from the past century are in rising of biomass and abundance of alien species (silver carp and perch) and domestic chub while on the other side we detected decreasing of biomass and abundance of nase, gray mullet and twaite-shad. In last several years in our research we haven't mullet and nase in our samples (MNE side), and this research is something that encourages us to state that this year sampling of those two species point toward kind of Lake recovery. Future monitoring have to give precise answer weather this was some normal natural circle or it was trend that we detected. Our standing point is that final answer and more precise characterization toward fulfilling of WFD needs could be given only after at least 5 years of the same standardized monitoring (MMG nets, electro-fishing transects, larval seining, long-lines and monitoring of commercial catch).

Table 19. Summarized fish species inventory for the Skadar/Shkodra Lake

Species	Native species (Literature)	Introduces Species (Literature)	Recorded under CSBL project sampling			
			MMG	Electro fishing	Commercial harvesting subsample	Commercial catch
Acipenser nacarii	Yes		No	No	No	No
Acipenser sturio	Yes		No	No	No	No
Alburnoides ohridanus	Yes		Yes	No	No	No
Alburnus scoranza	Yes		Yes	Yes	Yes	Yes
Alosa fallax	Yes		Yes	No	No	Yes
Ameiurus nebulosus		Yes	No	No	No	No
Anguilla anguilla	Yes		No	No	No	Yes
Atherina boyeri	Yes		No	No	No	No
Barbatula zetensis	Yes		No	No	No	No
Barbus rebeli	Yes		No	No	No	No
Carassius gibelio		Yes	Yes	Yes	Yes	Yes
Chondrostoma scodrensis	Yes		No	No	No	No
Chondrostoma ohridanus	Yes		Yes	Yes	No	No
Citharus linguatulus	Yes		No	No	No	No
Cobitis ohridana	Yes		Yes	No	No	No
Ctenopharyngodon idella		Yes	No	No	No	No
Cyprinus carpio	Yes		Yes	Yes	Yes	Yes
Dicentrarchus labrax	Yes		No	No	No	No
Gambusia holbrooki		Yes	No	No	No	No
Gasterosteus gymnurus	Yes		No	No	No	No
Gobio skadarensis	Yes		No	No	No	No
Hypophtalmichthys molitrix		Yes	No	No	No	No
Hypophtalmichthys nobilis		Yes	No	No	No	No
Liza ramada	Yes		No	No	No	No
Megalobrama terminalis		Yes	No	No	No	No
Mugil cephalus	Yes		Yes	Yes	No	Yes
Mylopharyngodon piceus		Yes	No	No	No	No
Oncorhynchus mykiss		Yes	No	No	No	No
Pachychilon pictum	Yes		Yes	No	No	No
Pelasgus minutus	Yes		No	No	No	No
Perca fluviatilis		Yes	Yes	Yes	Yes	No
Phoxinus limaireul	Yes		No	No	No	No
Pleuronectes flessus	Yes		No	No	No	No
Pomatoschistus montenegrensis	Yes		No	No	No	No
Pseudorasbora parva		Yes	Yes	No	No	No
Rhodeus amarus	Yes		Yes	No	No	No
Rutilus albus	Yes		Yes	No	No	No
Rutilus prespensis	Yes		Yes	Yes	Yes	Yes
Salaria fluviatilis	Yes		No	No	No	No
Salmo farioides	Yes		No	No	Yes	No
Salmo marmoratus	Yes		No	No	No	No
Salmothymus zetensis	Yes		No	No	No	No
Scardinius knezevici	Yes		Yes	Yes	Yes	No
Squalius platyceps	Yes		Yes	Yes	Yes	No
Telestes montenigrinus	Yes		No	No	No	No
Tinca tinca		Yes	No	No	No	No

Table 20. Skadar/Shkodra Lake fish species characterization table from the sampling campaign in autumn 2013/ winter 2014 (Presence/Absence in Reference period: L = evidenced by literature; C= evidenced by catch statistics; E = expert judgments; Abundance classes: 0 = absent, 1 = rare; 2 = frequent; 3 = abundant)

Species	Scientific name	Presence under reference (undisturbed conditions)	Presence today	Natural reproduction in lake	Abundance class under reference condition	Abundance class today	Comments
Adriatic sturgeon	*Acipenser nacarii*	L	L	Yes	1	0	It haven't been present in catch for almost 25 years
Atlantic sturgeon	*Acipenser sturio*	L	L	Yes	1	0	
Ohrid spirlin	*Alburnoides ohridanus*	L, C	L, E, C	Yes	3	2	
Bleak	*Alburnus scoranza*	L, C	L, E, C	Yes	3	3	
Twait shad	*Alosa fallax*	L, C	L, E	Yes	2	1	
Brown bullhead	*Ameiurus nebulosus*	-	L	Yes	-	0	
Eel	*Anguilla anguilla*	L	L, E	No	2	1	
Big-scale sand smelt	*Atherina boyeri*	L	L, E	No	1	1	
Zeta stone loach	*Barbatula zetensis*	L	L, E	No	1	1	
Western Balkan barbel	*Barbus rebeli*	L	L,E	Yes	2	1	
Prussian carp	*Carassius gibelio*	-	L, E	Yes	-	3	
Skadar nase	*Chondrostoma scodrensis*	L	L	No	1	0	
Ohrid nase	*Chondrostoma nasus*	L, C	L, E, C	No	3	1	
Atlantic spotted flounder	*Citharus linguatulus*	L	L, E	No	1	1	
Ohrid spined loach	*Cobitis ohridana*	L, C	L, E, C	Yes	1	2	
Grass carp	*Ctenopharyngodon idella*	-	L	No	-	0	Albania has stopped populating the lake since 1990, so their number is continuous decrease and probably will not be present in the lake in a short-term future
Carp	*Cyprinus carpio*	L, C	L, E, C	Yes	3	3	
See bass	*Dicentrarchus labrax*	L	L, E	No	1	1	
Mosquito fish	*Gambusia holbrooki*	-	L	Yes	-	1	
Western threespine stiklenek	*Gasterosteus gymnurus*	L	L, E	Yes	1	1	
Skadar gudeon	*Gobio skadarensis*	L	L, E	Yes	2	1	
Silver carp	*Hypophtalmichthys molitrix*	-	L	No	-	0	
Bighead carp	*Hypophtalmichthys nobilis*	-	L	No	-	0	

Thinlip mullet	*Liza ramada*	**L**	**L, E**	**No**	**1**	**1**	
Black Amur bream	*Megalobrama terminalis*	-	**L**	**No**	-	**0**	
Striped mullet	*Mugil cephalus*	**L, C**	**L, E, C**	**No**	**2**	**1**	
Black carp	*Mylopharyngodon piceus*	-	**L**	**No**	-	**0**	
Rainbow trout	*Oncorhynchus mykiss*	-	**L**	**No**	-	**0**	
Spoted roach	*Pachychilon pictum*	**L, C**	**L, E, C**	**Yes**	**3**	**3**	
Ohrid minnow	*Pelasgus minutus*	**L**	**L, E**	**Yes**	**1**	**1**	
Perch	*Perca fluviatilis*	-	**L, E, C**	**Yes**	-	**3**	
Adriatic minnow	*Phoxinus limaireul*	**L**	**L, E**	**Yes**	**3**	**2**	
Flounder	*Pleuronectes flessus*	**L**	**L, E**	**No**	**1**	**1**	
Skadar goby	*Pomatoschistus montenegrensis*	**L**	**L, E**	**No**	**1**	**1**	
Stone moroko	*Pseudorasbora parva*	-	**L, E, C**	**Yes**	-	**2**	
Bitterling	*Rhodeus amarus*	**L, C**	**L, E, C**	**Yes**	**2**	**1**	
White roach	*Rutilus albus*	**L, C**	**L, E, C**	**Yes**	**2**	**1**	
Roach	*Rutilus prespensis*	**L, C**	**L, E, C**	**Yes**	**3**	**3**	
Freshwater blenny	*Salaria fluviatilis*	**L**	**L, E**	**Yes**	**2**	**1**	
Adriatic trout	*Salmo farioides*	**L, C**	**L, E, C**	**No**	**2**	**1**	
Marble trout	*Salmo marmoratus*	**L**	**L, E**	**No**	**1**	**1**	
Soft-muzzled trout	*Salmothymus zetensis*	**L**	**L**	**No**	**1**	**0**	
Skadar rudd	*Scardinius knezevici*	**L, C**	**L, E, C**	**Yes**	**3**	**2**	
Shub	*Squalius platyceps*	**L, C**	**L, E, C**	**Yes**	**3**	**2**	
Montenegro riffle dace	*Telestes montenigrinus*	**L**	**L, E**	**Yes**	**3**	**2**	
Tench	*Tinca tinca*	-	**L**	**Yes**	-	**0**	

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8. Annex

8.1. Annex 1 – Length/Weight scatter plots and growing curves based on electrofishing samples

Regarding insufficient number of species individuals in pulled samples from all three transects we haven't construct length frequency graph that shows population structure in sample. However species individuals in sample were sufficient for constructing of growing curve what we couldn't do based on MMG gillnet samples (larger sized fish species: carp, chub, nase and Prussian carp).

Cyprinus carpio **(carp)**

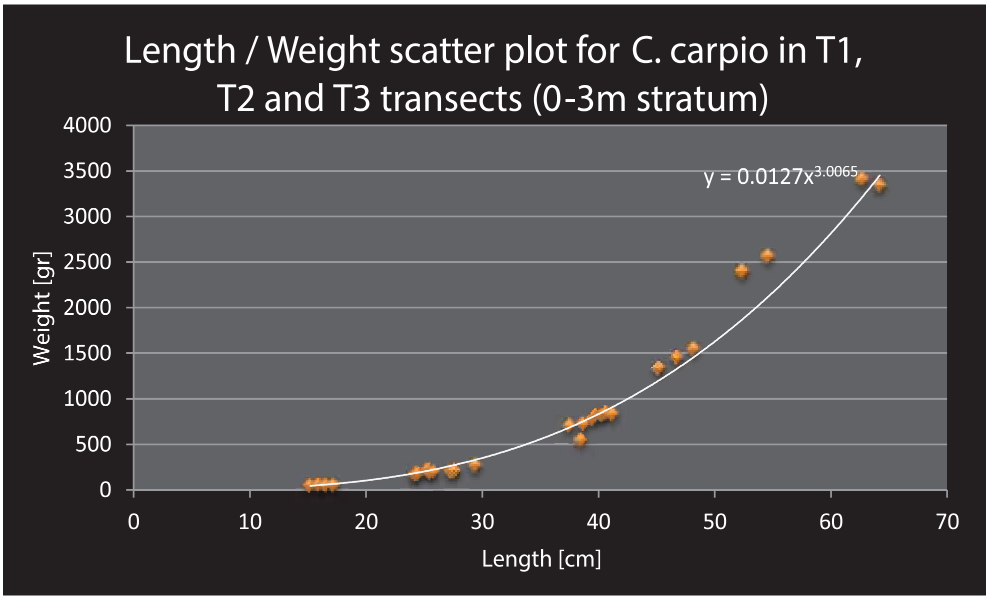

Figure 1. Length/Weight scatter plot and growing curve for carp (C. carpio) in 0-3m stratum (electrofishing transect, T1, T2 and T3)

Based on constructed growing curve we can conclude that carp has isometric growing pattern in 0-3m water mass of Skadar/Shkodra Lake. Although it was reported that carp has isometric growing (Prpa et al., 2007, MIlosević & Marić, 2012) our finding could be consequence of low number of individuals in sample, especially those of middle and large body size and future sampling and analyzing of pulled samples will give us precise information about carp growing pattern.

Carassius gibelio **(Prussian carp)**

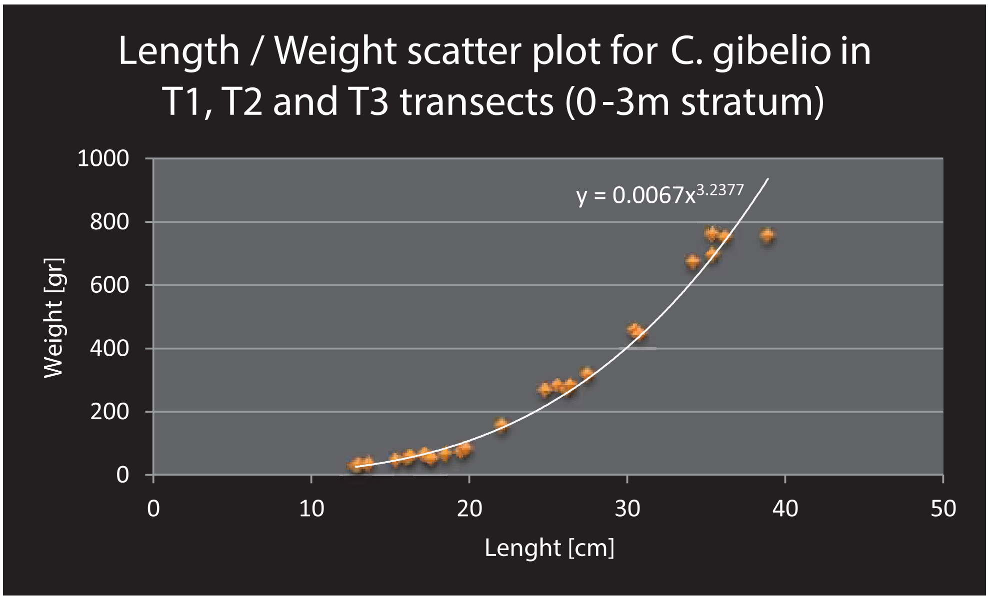

Figure 2. Length/Weight scatter plot and growing curve for Prussian carp (C. gibelio) in 0-3m stratum (electrofishing transect, T1, T2 and T3)

Based on constructed growing curve we can conclude that Prussian carp has positive allometric growing pattern in 0-3m water mass of Skadar/Shkodra Lake.

Squalius platyceps (**chub**)

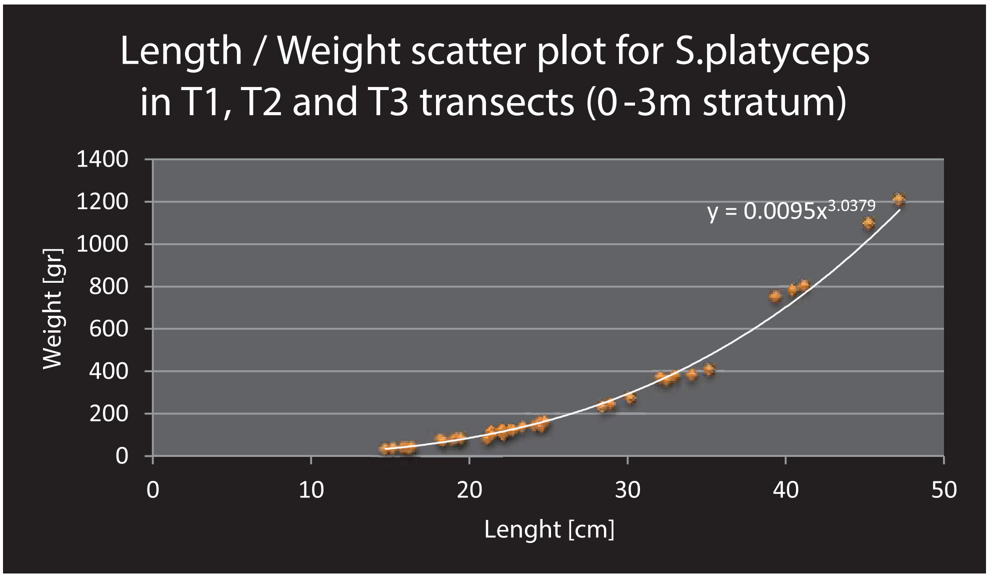

Figure 3. Length/Weight scatter plot and growing curve for chub (S. platyceps) in 0-3m stratum (electrofishing transect, T1, T2 and T3).

Based on constructed growing curve we can conclude that chub has isometric growing pattern in 0-3m water mass of Skadar/Shkodra Lake. Our finding could be consequence of low number of individuals in sample, especially those of middle and large body size and future sampling and future analyzing of pulled samples will give us precise information about chub growing pattern.

Chondrostoma nasus (**nase**)

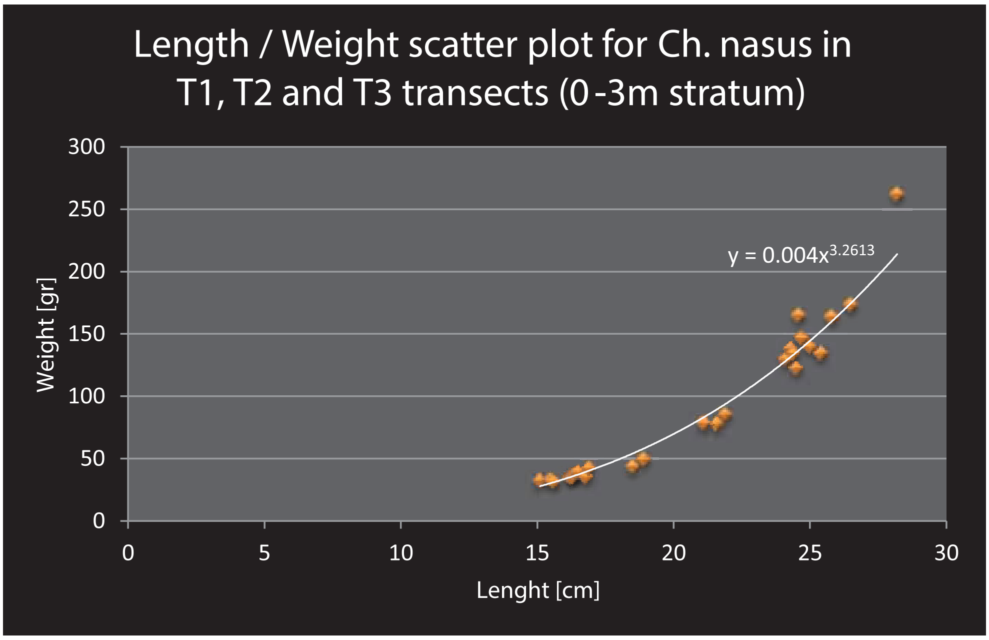

Figure 4. Length/Weight scatter plot and growing curve for nase (Ch. nasus) in 0-3m stratum (electrofishing transect, T1, T2 and T3).

Based on constructed growing curve we can conclude that nase has allometric growing pattern in 0-3m water mass of Skadar/Shkodra Lake. Our finding could be consequence of low number of individuals in sample and future analyzing of pulled samples will give us precise information about nase growing pattern.

REPORT

PHYSICO-CHEMICAL INVESTIGATIONS OF THE WATER OF LAKE OHRID AND MAIN TRIBUTARIES

Trans-boundary Monitoring program

Prepared by Dr Elizabeta Veljanoska-Sarafiloska
Hydrobiological Institute, Ohrid, R. Macedonia

Contents

List of abbreviations

BOD_5 - biochemical oxygen demand
NO_2-N - nitrite-nitrogen
NO_3 - nitrate nitrogen
NH_3-N - ammonia-nitrogen
$TN_{Kjeldahl}$ - total nitrogen by Kjeldahl
TN - Total nitrogen
TP - total phosphorus
TSI - trophic state index
OCP - organochlorine pesticides

1. Introduction

The ecological system of lakes is highly dependent on the environment, i.e. on the physical, chemical and biological conditions in the watershed area. Human impact and the changes caused by human activities as well as natural changes could cause an increasing flow of nutrients. Nutrients especially phosphorus as limiting element of euthrophication can affect of trophic state of the water bodies. Ohrid and Prespa Lake belong to the largest and most important natural water ecosystems in the Balcan region. They belong to the Desaretian group of lakes and with their natural rarities and endemic species.

The classification of the water bodies is made according the Carlson index for trophic state (1977), OECD clasification (1982) and Directive for Classification of Waters (1999)
The application of the Carlson's method for determination of the trophic state of water considers the following parameters:

- Concentration of total phosphorus
- Secchi transparency

2. Parameter and sampling points

Water samples were collected from following sampling sites on the lake:

MK I Kalishta (N 41°09'025 E 20°39'078),

MK II Grashnica (N 41°07'043 E 20°47'260),

MK III Velidab (N 40°59'237 E 20°47'965),

MK IV St. Naum (N 40°54'855 E 20°44'499),

MK V Pelagic zone (N 41°03'730 E 20°45'000)

Additionally water samples were collected from the following sampling sites at the main tributaries:

Sat 1 River Sateska before redirection (N 41°13'569 E 20°44'755),

Sat 2 River Sateska middle course (N 41°12'959 E 20°44'811),

Sat 3 River Sateska inlet (N41°10'071 E 20°43'620),

Kos 1 River Koselska (N41°07'355 E 20°46'329),

Che 1 River Cerava inlet (N 40°55'402 E 20°45'376)

Sediment samples were collected from following sampling sites: MK V (St. Naum) and MK II (Grashnica)

Figure 1: Sampling sites Lake Ohrid pelagic zone, littoral zone and main tributaries

Following physico-chemical parameters have been analysed within the investigations of the CSBL project:

- transparency
- temperature
- pH
- conductivity
- dissolved oxygen
- biochemical oxygen demand (BOD_5)
- organic matter (through permanganate consumption)
- oxygen saturation
- total phosphorus
- nitrite-nitrogen (NO_2-N)
- nitrate-nitrogen (NO_3-N)
- ammonia-nitrogen (NH_3-N)
- $TN_{Kjeldahl}$ (total nitrogen by Kjeldahl)
- TN total nitrogen
- suspended solids

3. Description of investigations

The sampling of water at all measuring stations, was carried out in four campaigns, which are belonged to four different seasons as recommended in the WFD: April, July, October and January.

Samples for physico-chemical parameters have been taken during four sampling campaigns:

- first campaign in spring period 2013, the weather in this campaign was sunny with a light south wind
- second campaign in the summer period 2013, the weather in this campaign was sunny
- third campaign in the autumn period 2013, the weather in this campaign was sunny
- fourth campaign in the winter period 2014, the weather in this campaign was snow, wind with cloudy

Collecting of samples from pelagic was done by boat belonging to the HBIO. Two days were spent for collection of samples in each campaign (one for pelagic and one for rivers and littoral zone).

Ruttner bottle of 2,25 l (Hydro-bios, Kiel, Germany) was used for collecting the water samples from the littoral zone. The water samples from the pelagic zone (at two depths down the water column: 1 and 15 m), were collected with Niskin bottle (5l). The water sample from the river was collected directly into bottles. All sampling procedures fullfilled the requirements of official standard methodologies.

Samples for general chemical analysis were collected in polyethylene bottles. Samples for oxygen and BOD_5 were collected in Winkler bottles of 110-140ml and add 0.5 mL $MnSO_4$ followed by 0.5 mL KJ immediately after the sample collected. Samples for total phosphorus were collected in 130 ml polyethylene bottles and were filled completely with sample after rinsing them twice with the water to be analysed.

Samples for nitrogen compound (nitrite, nitrate, ammonia), should be collected in polyethylene bottles (500 ml) and were analysed immediately within 24h after collection. The best technique in nitrogen analysis is to do any filtrations within 5-10 hours of taking samples (which are kept cool and dark) and then quickly deep-freeze filtrates to -20 ^{o}C in polyethylene bottles.

Sediment samples were collected at the two sampling points Otesevo littoral and Ezerani littoral. The sampling was conducted with a Van-Veen grab sampler with a volume of 440 cm^3 (Hydro-bios, Kiel, Germany).

4. Transport and storage of samples

Bottles with samples were disposed in hand refrigerators at low temperature. Samples were transported to laboratory by car. The containers with samples were saved of any outside influences. Samples were transported to laboratory immediately. After their arrival in the laboratory they were disposed in refrigerators at 4^{o}C till the analysis. Samples for total phosphorus should be kept in a cool dark place (4 ^{o}C) and not warmed to room temperature until the analysis has been to be commenced.
Sediment samples were stored and transported in field – refrigerators. Before analysis samples were stored at 4 ^{o}C for a maximum of seven days.

5. Analysis

All analyses have been done by standard methods (APHA-AWWA-WPCF 1980).

Transparency was measured by Secchi disk (secchi disk is a 20 cm diameter). Temperature was measured by reversing thermometer (Welch, 1948), pH was measured by WTW, Multilab 540 and conductivity by WTW conductometer LF 197. Temperature of water, transparency by Secchi disk, pH and conductivity were measured on the field.

Alkalinity was determined by titrating with standard sulphuric acid (0.02N) at room temperature using phenolphthalein and methyl orange indicator).

The organic matter content in the water expressed as permanganate consumption was determined by digestion in acid and by titration (Bether,1953).

Winkler method was used for analysis of dissolved oxygen and BOD_5. The fixation of dissolved oxygen was immediately after the sample collected in a winkler bottle with 0.5 mL $MnSO_4$ followed by 0.5 mL KJ.

Standard limnological methods (Beter, 1953; Ruttner, 1975; Golterman et al., 1978; Wetzel & Likens, 2000; APHA-AWWA-WPCF, 2005)were used. for all titrimetric analyses.

The *Total Organic Carbon* (TOC) (index of the total amount of organic substances in water) was measured using a Total Organic Carbon Analyzer TOC – TOC ID.

Method 2540D in *Standard Methods for the Examination of Waters and Wastewaters,* 20th Edition (American Public Health Association, 1998) was used for total suspended solids. Therfore a glass-fiber filter retains the suspended solids. After filtering the sample, the filter is dried to a constant weight at 103–105 °C.

Nitrates are quantitatively reduced from the water to nitrites by a cadmium-cooper couple (Stricland & Parsons, 1972; APHA-AWWA-WPCF 1980).

The procedure for detemination of ammonia is performed by binding of the ammonia with hypohlorite in monochromine which in reaction with phenol gives p-aminophenol. In reaction with natrium nitroprusside it forms blue-coloured compounds (Solorzano 1969., Wetzel and Likens 1979). Water for ammonia nitrogen determination should be filtered.

Total nitrogen is the sum of organic nitrogen and inorganic nitrogen. In principle organic nitrogen but not nitrate or nitrite is reduced to ammonium during Kjeldahl digestion. The determination of ammonium in Kjeldahl digestion measures organic nitrogen + ammonium.

$$TN = \text{organic ntrogen} + \text{ammonium} + \text{nitrite} + \text{nitrate}$$

Kjeldahl digestion method (Strickland, J. D. H., Parsons, T.R., 1968; APHA-AWWA-WPCF, 1980) was used for dermination of total nitrogen. The results are read on using Perkin-Elmer UV-VIS, spectrophotometer, wavelenght 530 nm (nitrite and nitrate) and 640 nm (ammonia and total nitrogen by Kjeldahl).

Total phosphorus was analyzed by acid digestion of persulphate (at 121 °C, pressure 1 at, and time 1 hour); all forms of phosphates are altered to orthophosphate (Strickland and Parsons 1968; Menzel & Corwing, 1965; ISO/DIS 6878/1 : 1984).

The analysis for orthophosphate has been performed using the filtered water without acid digestion (mineralization before the chemical analysis). 130 ml polyethylene bottles should be filled completely with sample after rinsing them twice with the water to be analysed. The analysis should be commenced as soon as possible, perferably within 1/2 hr, certnly before 2 hr. (Strickland and Parsons 1968; ISO/DIS 6878/1 : 1984).

The results for total phosphorus and ortophosphate is read on Spectrophotometer Specord; model S-10, Carl Zeiss Jena, 880 nm.

The water for some parameters (nitrogen compounds, ortophosphate) were filtered through the glass filter 0,45µm, as soon as they come in laboratory.

The determination of organochlorine pesticides in the sediment was conducted using the modified EPA 8081A method. 70 to 80 g of fresh and well-homogenized sediment were placed in a glass container and mixed with a 1:1 mixture of hexane and acetone (*V*/*V*) and of methanol. Solid-liquid extraction was performed on a magnetic stirrer for 2 hours at room temperature without any thermal treatment of the sample.

Quantitative analysis for organochlorine pesticides was conducted with a gas chromatograph (Varian, USA) Model 3800, with an ECD detector and nitrogen as the carrier gas. The column used for separation of OCP was VA-1701 (VA-123073-20) with length 30 m; I.D. 0.32 mm; film thickness 0.25 mm, and temperature limits from − 20 °C to 280 °C (300 °C).

Classification of the water has been done according Carlson index for trophic state (1977), OECD clasification (1982) and Directive for Classification of waters (1999)

The application of the Carlson's method for determination of the trophic state of water which refers to the following parameters: concentration of total phosphorus and Secchi transparency, provides the numeric value of the trophic state index.

The classification of the obtained numerical values of the indexes is according trophic scale proposed by Aizaki (1981).

6. Evaluation

6.1 Lake Ohrid pelagic zone

Transparency (s.c. Secchi depth)

Transparency of Lake Ohrid water column varied between 15.5 m during winter period and 21 m during autumn.

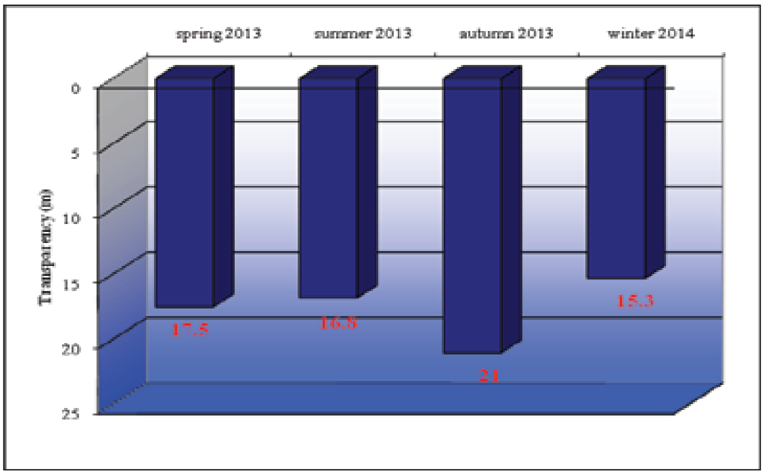

Figure 2: Transparency (Secchi depth) changes of Lake Ohrid

Temperature

Water temperature in Lake Ohrid typically varies from the surface to the deeper layers. In general, temperatur increase in the upper water column starts during the heating period (spring and summer months). Figure 3 shows typical stratification in the pelagic zone of Lake Ohrid during the summer and spring period. Temperature of the pelagic water column varied between 6.09 °C at depth of 240 m, almost all investigated period, and 23.22 °C at the surface water during the summer period.

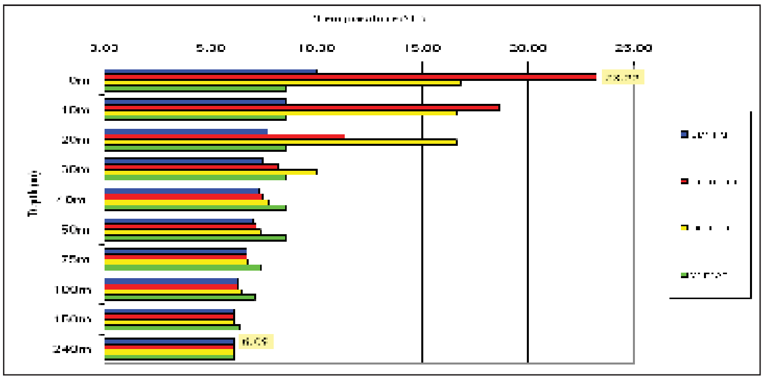

Figure 3: Temperature changes in the water column of the pelagic zone of Lake Ohrid

According the mictic characteristics Lake Ohrid can be classified as an oligomictic lake according Hutchinson and Loffler (1956) and cold oligomictic sensu Hadzisce (1966).

<u>Conductivity</u>

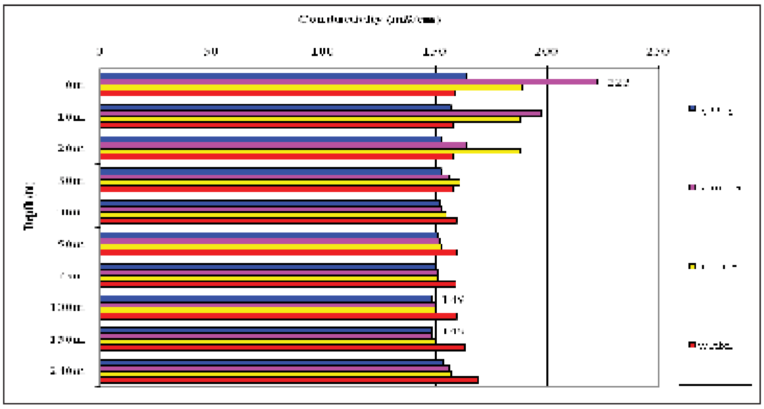

Figure 4: Conductivity changes in the water column of the pelagic zone of Lake Ohrid

Conductivity in the water column of Lake Ohrid in the pelagic zone has varied between 149 mS cm^{-1} at 100 and 150 m depth during a longer investigated period (spring, summer and autumn) to 223 mS cm^{-1} at the surface water during the summer. As it can seen bigger changes are happening in the upper layer of the trophogenic zone of lake Ohrid.

<u>pH</u>

pH value is close connected to the lake Ohrid metabolism. pH values for this parameter have varied between 7.92 at 240 m depth during the spring period and 8.87 at 20 m during the summer period. pH values as a result of the metabolic activities were higher in the thermocline of the pelagic zone of Lake Ohrid.

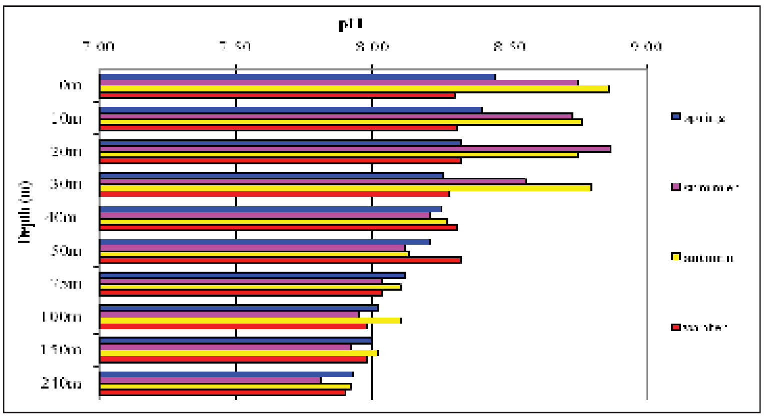

Figure 5: pH changes in the water column of the pelagic zone of lake Ohrid

Total alkalinity

Total alkalinity values have varied from 106 mg l^{-1} $CaCO_3$ at surface water (autumn), 30 and 50 m (summer) to 121 mg l^{-1} $CaCO_3$ at 240 m depth during summer period (Fig. 6).

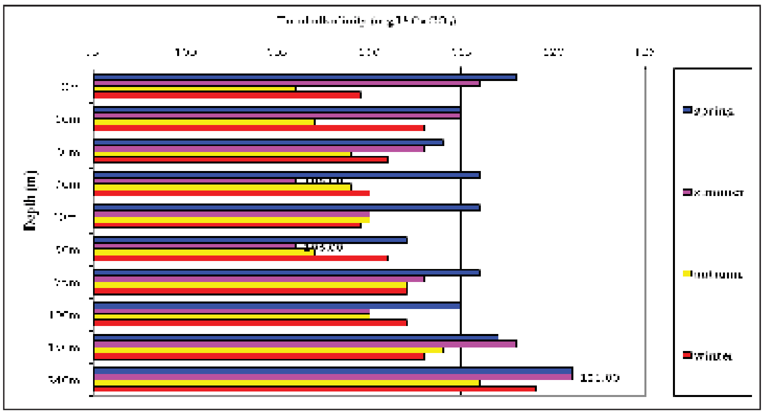

Figure 6: Total alcalinity changes in the water column of the pelagic zone of Lake Ohrid

Dissolved oxygen

Dissolved oxygen in the water column of a lake is a key substance in its metabolism. It is a result of the biochemical processes that occur continuosly by variable intensity.

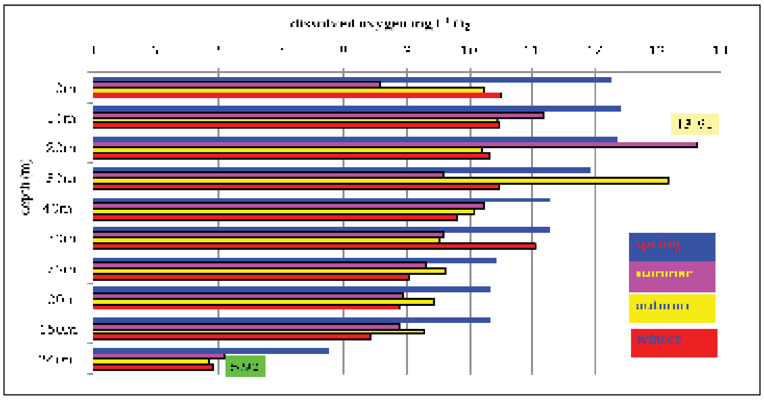

Figure 7: Concentration of dissolved oxygen in the water column of the pelagic zone of Lake Ohrid

During all sampling campaigns it was observed that all depths have been well supplied with oxygen. The highest concentration of dissolved oxygen (13.61 mg l^{-1} O_2) was measured in the trophogenic zone at 20 m depth during summer period. The lowest concentration (5.92 mg l^{-1} O_2) was measured at the lowest part of the tropholitic zone at 240 m depth. During the winter period concentration for dissolved oxygen are very close whichis the result of the circulation process within the water body. No anoxic condition are registered in any part of Lake Ohrid. Lake Ohrid according to this parameter has an oligotrophic status (Directive for Clasiffication of Waters).

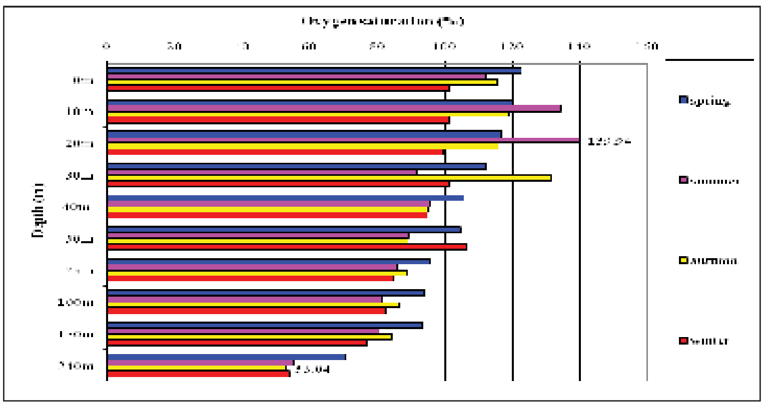

Figure 8: Oxygen saturation changes in the water column of the pelagic zone of Lake Ohrid

Oxygen saturation

Oxygen saturation in the water column of Lake Ohrid indicates high oxygen saturation in the trophogenic zone with a maximum of 139 % at 20 m depth during summer. Minimal oxygen saturation was measured in the lower tropholitic zone (on which mineralization processes prevail) at depth of 240 m during autumn.

Oxygen released during photosynthesis is used for respiration of all the lake biota and oxidation of numerous biodegradable substances. A suitable method to measure such substances is the permanganate consumption. Higher permanganate consumption is characteristic for the trophogenic zone in which, because of intensified physiological processes more biodegradable substances are released by excretion or not completed food ingestion. Concentration of organic matter at the pelagic zone are generaly below 5 mg l^{-1}. Thus lower permanganate consumption was found in the deeper water layers of the water column (2.34 mg l^{-1} O_2 at 100 and 150 m depth during summer). Highest value for this parameter was registred during winter period at the surface water (4.48 mg l^{-1} O_2) and 40 m (4.81 mg l^{-1} O_2) depth.

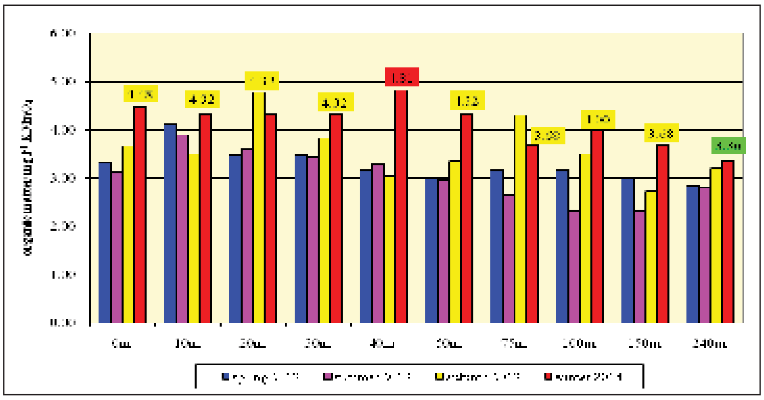

Figure 9: Permanganate consumption changes in the water column of the pelagic zone of Lake Ohrid

All values in the winter period were very close (from 3.36 on 240 m to 4.81 mg l^{-1} on 40 meters. According of Directive for Clasiffication of Waters, Lake Ohrid pelagic zone based on this parameter is in I-II class.

Biochemical oxygen demand (BOD5)

BOD_5 is the amount of dissolved oxygen needed by aerobic biological organisms to break down organic material present in a given water sample at certain temperature over a specific time period. The term also refers to a chemical procedure for the determination of this amount. This is not a precise quantitative test although it is widely used as an indicator of the organic quality of water.The BOD value is most commonly expressed in milligrams of oxygen consumed per litre of sample during 5 days of incubation at 20 °C and is often used as a robust surrogate of the degree of organic pollution of water.

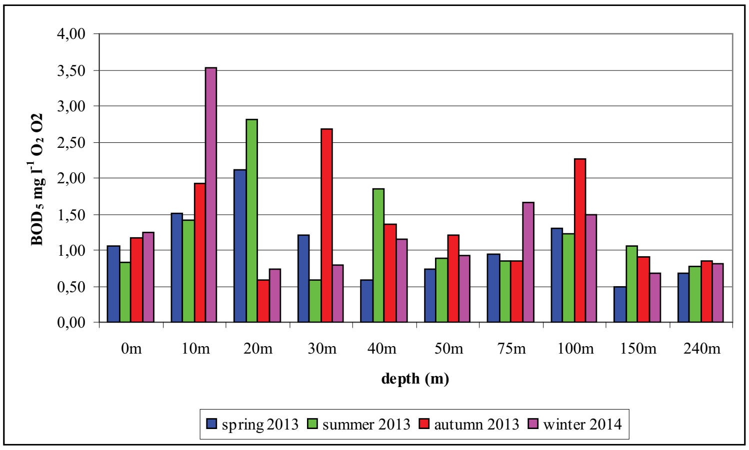

Figure 10: Biochemical oxygen demand within five days (BOD_5) changes in the water column of the pelagic zone of Lake Ohrid.

Higher values were found in the termocline, the upper layer of the trophogenic zone. The highest BOD_5 value, 3.52 mg l^{-1} O_2 was registered at 10 m depth during winter, lowest value, 0.67 was at 240 m depth during spring period.

The BOD_5 values indicate a low presence of dissolved biodegradable organic matter in the water column of Lake Ohrid and low density of aquatic organisms (phytoplankton, zooplankton and bacterioplankton), which is a significant characteristic of Lake Ohrid as an oligotrophic lake.

Total phosphorus

Total phosphorous as an essential nutrient is the most limiting factor in the eutrophication processes of aquatic ecosystems. Even a small amount of phosphorus to a water body can have negative imapct for the water quality. Those adverse effects include: algae blooms, accelerated plant growth, and low dissolved oxygen from the decomposition of additional vegetation.

Measuring concentration of total phosphorus are generaly below 10 mg l^{-1}. On the deepest point, near the sediment is the highest concentration for total phosphorous (13,88 mg l^{-1}). The lowest concentration has been measured at 100 m depth during the autumn (2.11mg l^{-1}).

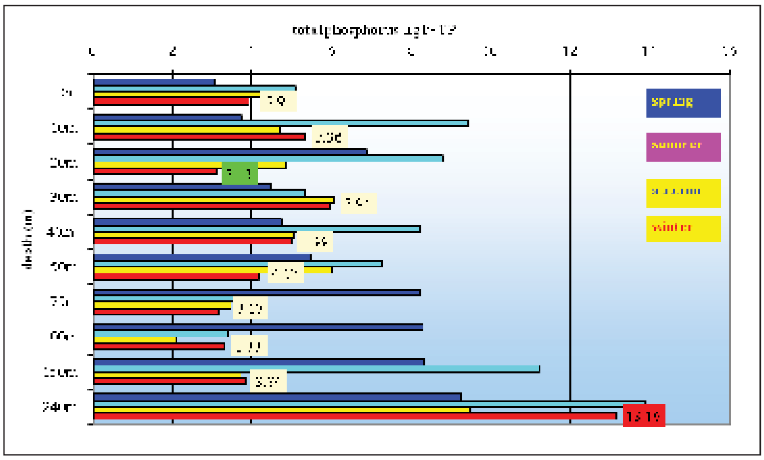

Figure 11: Total phosphorus concentration changes in the water column of the pelagic zone of Lake Ohrid

Main phosphorus loads to Lake Ohrid originate from land wash out through the numerous tributaries and probably from sublacustrine springs that feed Lake Ohrid.

According to the OECD classification and Directive for Classification of Water based on concentration of total phosphorus, Lake Ohrid belong to **oligotrophic state, ie I class** of water.

Nitrogen compounds

Nitrogen is another relevant nutrient according to the eutrophication process. It can be present in the lake water in form of nitrite, nitrate, ammonnia and total nitrogen by Kjeldahl (organic nitrogen). Tributaries are the biggest sources of nitrogen loads of Lake Ohrid as well. However contrary to phosphorus nitrogen compounds can be supplied from the atmosphere as well as from tributaries. Usually nitrogen is second limiting nutrient in the productivity and eutrophication of aquatic ecosystems.

Because of well - oxygenated water in the water column in the pelagic zone of Lake Ohrid, concentrations of nitrite-nitrogen and ammonia-nitrogen in the water column have been very low, (below 0.001 mg l^{-1}.

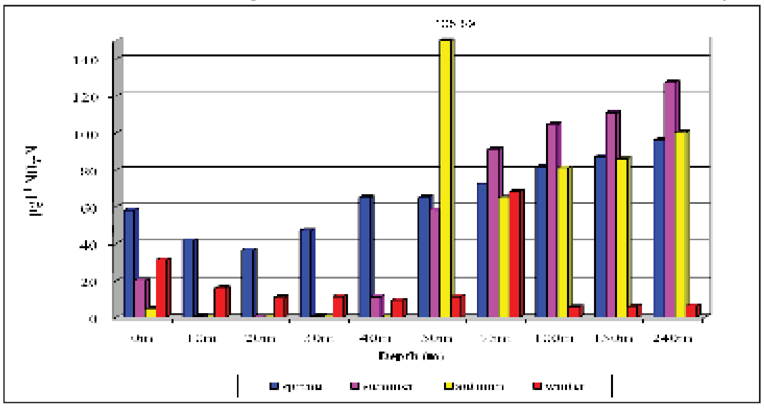

Figure 12: Nitrate nitrogen concentration in the water column of the pelagic zone of Lake Ohrid.

Probably because of the fast uptake of the phytoplankton community concentrations of nitrate-nitrogen in the upper layer of the trophogenic zone were negligible. Higher values were measured at deeper layer from the lake. The highest nitrate-nitrogen concentration in the water column of Lake Ohrid was found at 50 m depth during the autumn (405.89µg l^{-1}).

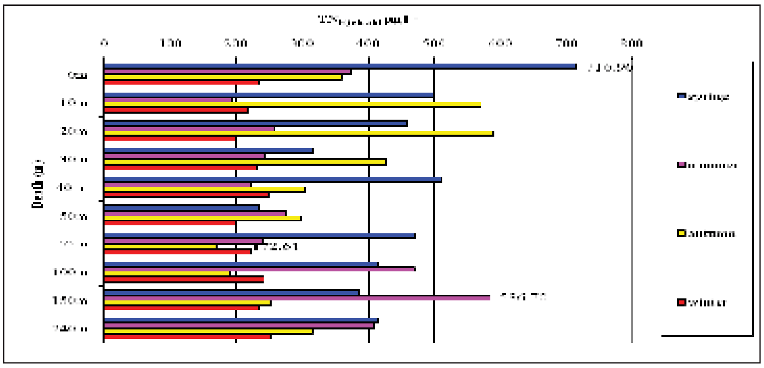

Figure 13: Total Nitrogen by Kjeldahl concentrations in the water column of the pelagic zone of Lake Ohrid

Concentrations of the total nitrogen by Kjeldahl in the water column of Lake Ohrid are very variable. However it is a presence of two maximums at two different depths (surface water during the spring - 716.96 mg l^{-1} and 150 m depth during the summer 586.70 mg l^{-1}). The lowest value for total nitrogen by Kjeldahl was measured at 75 m depth during autumn, 172.61mg l^{-1}.

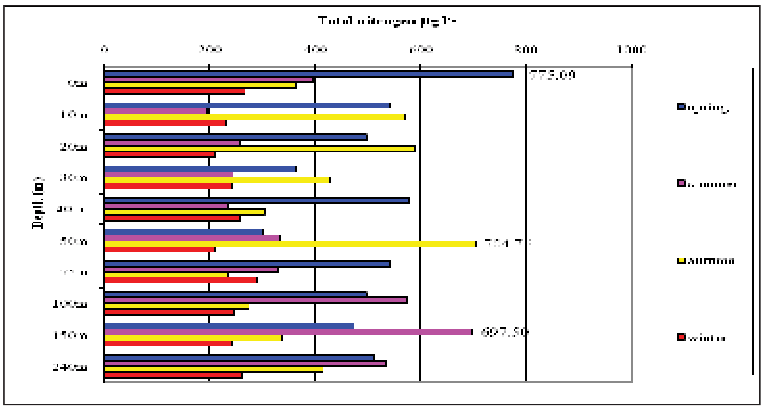

Figure 14: Total nitrogen concentration in the water column of the pelagic zone of Lake Ohrid

Highest values for total nitrogen are registered at surface water (775.09 mg l^{-1}) during the spring period, at 50 m depth during the autumn (704.76mg l^{-1}) and at 150 m depth during the summer period (697.5 mg l^{-1}). Concentrations of the total nitrogen by Kjeldahl in the water column of Lake Ohrid are very variable but usually are below 400 mg l^{-1}. Higher values were measured at the surface layer and at the deeper layer of the lake. Antropogenic impacts as well as atmospheric deposits (for the surface water) and more intensive process of decomposition of organic matter in the deeper layers of the lake are probable causes. According to the OECD classification and Directive for Classifications of Water (Official Gazette, of R.M., N° 18/99) Lake Ohrid pelagic zone has still a oligotrophic character.

6. 2. Lake Ohrid littoral zone and main tributaries

Temperature

Highest values were measured during the summer period for all investigated points (23.8 °C Veli dab littoral). The lowest value for this parameter was measured in River Sateska (6.1 °C, winter period).

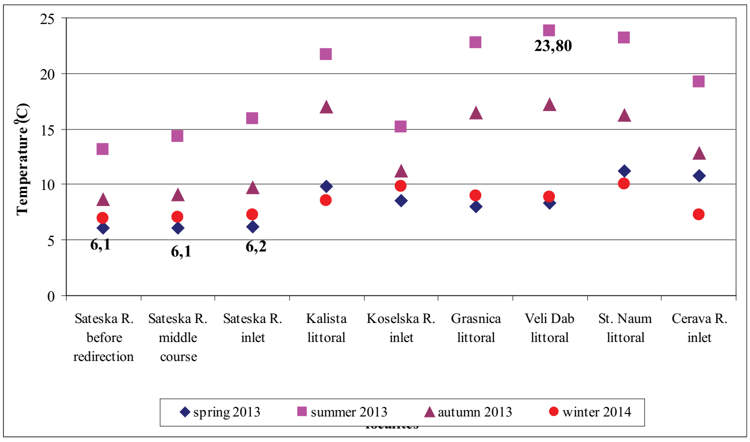

Figure 15: Temperature changes in the water from sampling points

pH

Lowest values for pH for all investigated period were registered in water samples collected from River Koselska inlet (7.59 during spring).

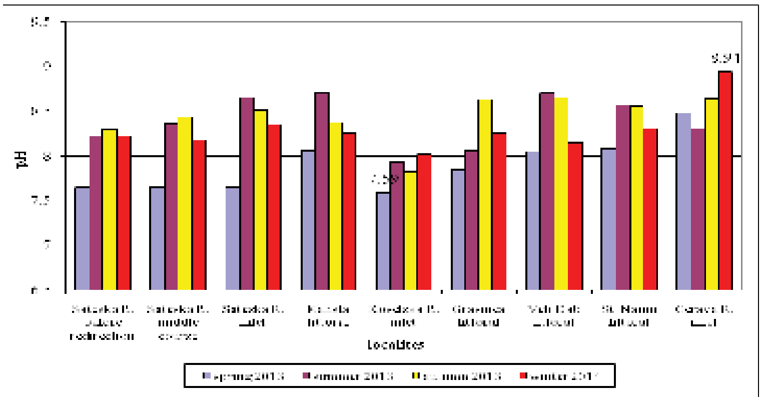

Figure 16: pH changes in the water from sampling points

Maximal value was registered at Cerava River inlet during winter period (8.94).

Conductivity

Conductivity in the water from sampling points varied between 102 mS cm^{-1} at Grasnica littoral (spring period) and 439 mS cm^{-1} during summer and autumn (Fig. 17)

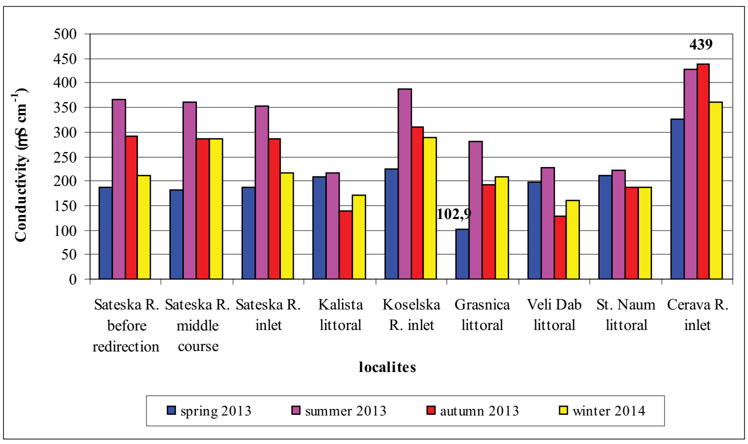

Figure 17: Conductivity changes in the water from sampling points

Total alkalinity

Total alkalinity values varied from 110 mg l^{-1} $CaCO_3$ at Kalista littoral (autumn), 208 mg l^{-1} $CaCO_3$ at Cerava River inlet during autumn period.

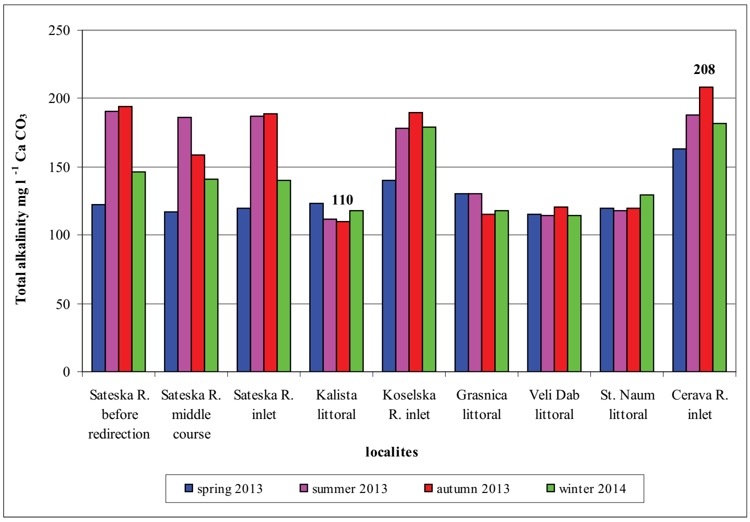

Figure 18: Total alkalinity changes in the water from sampling points

Total suspended solids

The Minimum values for total suspended solids during the investigated period were registered in the the littoral zone of the Lake (0.012 g l-1 at St. Naum and 0.014 g l-1 at Veli Dab during the autumn).

The maximum values in all analysed periods, for the total suspended solids were registered in River Koselska's inlet. In the summer period 0.221 g l-1 were registered for this parameter . In River Cerava's inlet higher values for total suspended solids during the investigated period were registered as well. The maximum values from these sampling points were measured in the summer period, 0.258 g l-1.

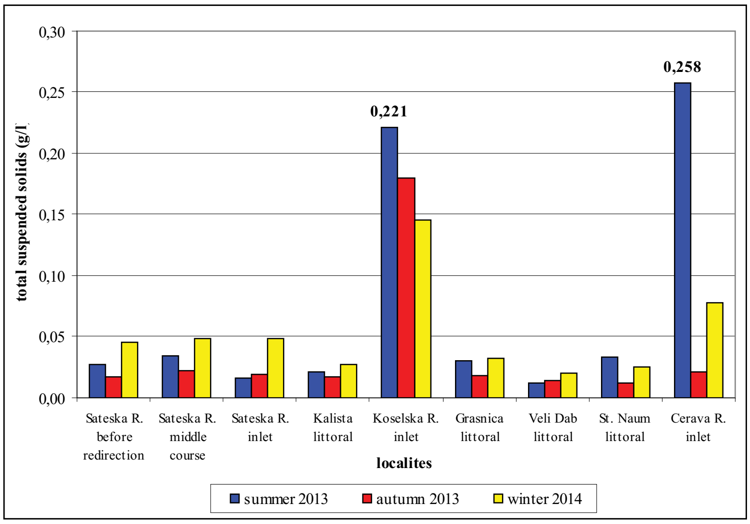

Figure 19. Total suspendet solids in the water from the sampling points

Dissolved oxygen

Oxygen is essential to the production and support of all life in the lakes i.e in all water ecosystems.

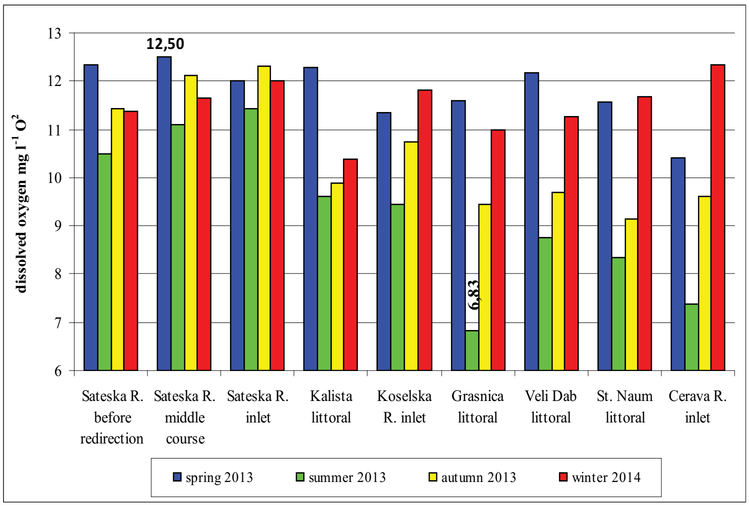

Figure 20: Concentration of dissolved oxygen

During the whole investigated period (four campaigns) all sampling points are good supplied with oxygen. All values for this parameter are above 8 mg l^{-1} O_2. Exception of this situation is observed in localities Grasnica littoral and River Cerava inlet during the summer campaign, when were measured the lowest values for this parameter (6.83 for littoral Grasnica and 7.38 for R. Cerava inlet).

Oxygen saturation

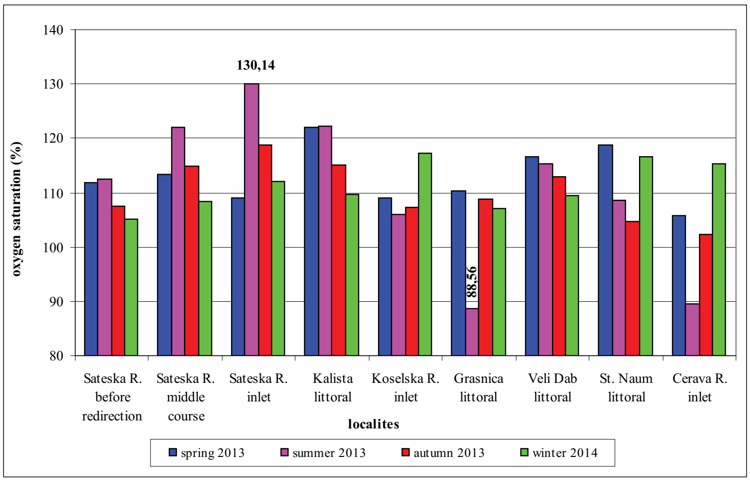

Figure 21: Oxygen saturation in the water from sampling points

Oxygen saturation in the water samples from littoral zone and tributaries indicates a high oxygen saturation with a maximum of 130.14 % at Sateska River inlet during summer period. The lowest oxygen saturation was measured in the Grasnica littoral during summer (88.56 %).

Biochemical oxygen demand

The required oxygen for biochemical degradation of organic components in combination with microorganisms is expressed through biochemical oxygen demand for five days (BOD_5).

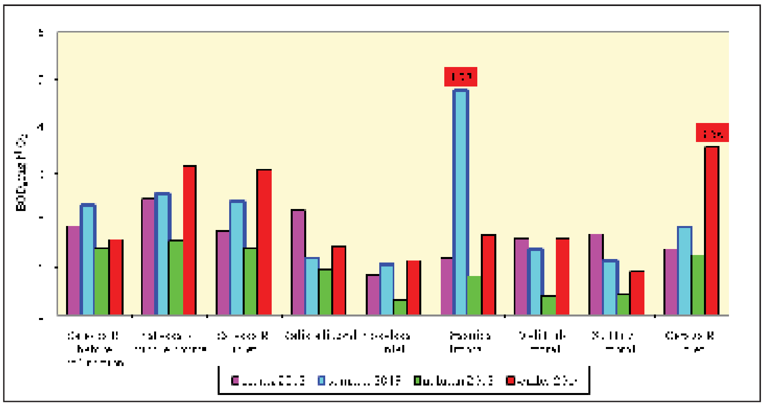

Figure 22: Biochemical oxygen demand in water from sampling point

According to the obtained results highest values for BOD were measured in all sampling points during the summer and winter period. Maximum value during the summer period is measured in littoral Grasnica (4.77 mg l^{-1} O_2). The highest value during the winter period was registered in Cerava R. inlet (3.56 mg l^{-1} O_2). The highest value of BOD_5 has been registered in the littoral Grasnica, presumably as a reflection of the influence from the River Vegoska which delta is nearby. Likewise relatively high values of this indicator have been registered in River Sateska middle course and River Sateska inlet during the summer and winter period.

The lowest value for biochemical oxygen demand (BOD_5) during all investigated period have been measured at St. Naum littoral zone.

Organic matter

The consumption of $KMnO_4$ is an indirect measure for the quantity of organic biodegradable matter in the water (the quantity of consumed permanganate depends on the quantity of organic substance in the water and their chemical structure).

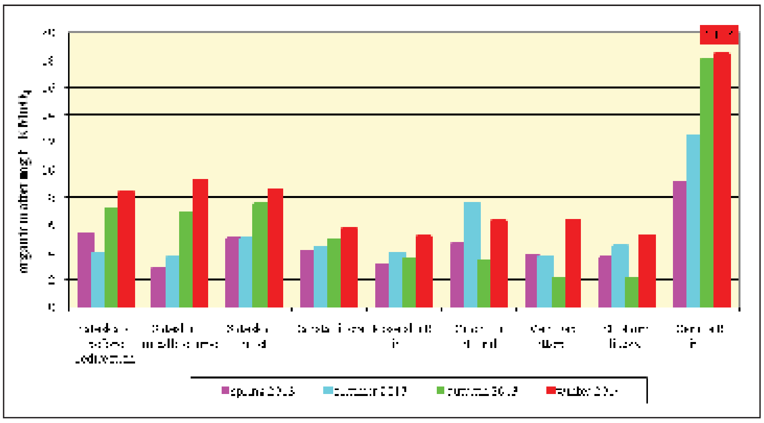

Figure 23: Organic biodegradable matter in water from sampling point

The inlet of River Cerava had obviously the highest values during all sampling campaign with a maximum of 18.44 mg l^{-1}. During the summer period the highest values for content of organic matter are evident in water samples from littoral Grasnica and River Cerava inlet. Likewise relatively high values of this indicator have been registered in River Sateska. This situation is due to the fact that these rivers pass through agricultural areas and settlements and they are "the end-recipients" of the industrial waste water, drainage water and sewage from households which directly flowing into the lake littoral.

Nitrogen compounds

The nutrient loading of the water from investigated points has been determined based on the concentration of two most important biogenic elements: total phosphorus and total nitrogen.

There were analyzed several nitrogen compounds present in the water samples as: NH_3-N, NO_2-N, NO_3-N, TNKjeldahl, and total nitrogen. These nitrogen compounds take part in different physiological processes they directly depend of the processes of production and mineralization which parallel exist in the water.

Nitrite-nitrogen

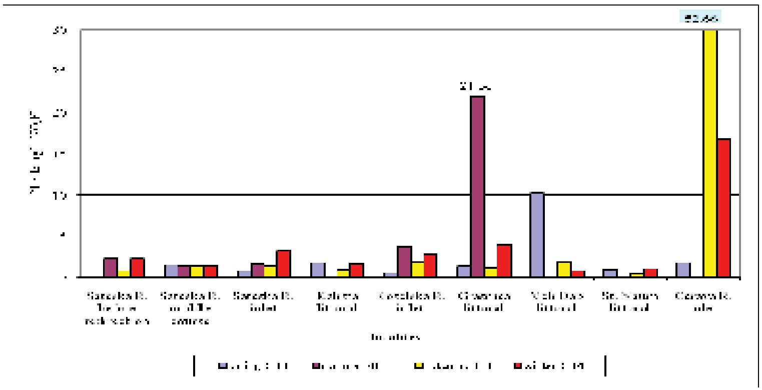

Figure 24: Nitrite - nitrogen concentration in water from sampling points

Concentration for this parameter have been very low during the all investigated periods. Maximal values were determined in Cerava River inlet (52.66 mg l^{-1}) during autumn and in Grasnica littoral 21.88 mg l^{-1} during summer.

Nitrate

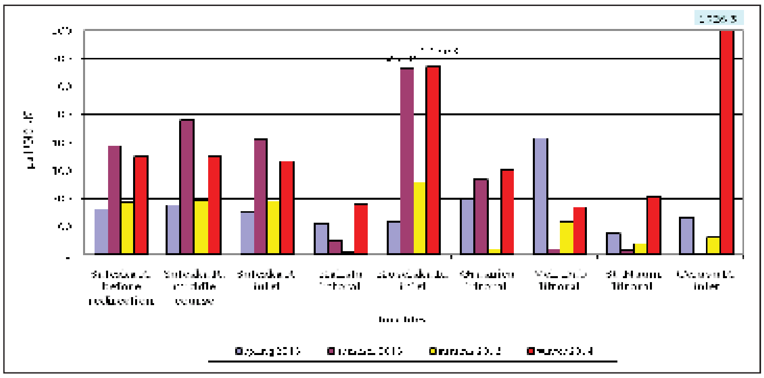

Figure 25: Nitrate - nitrogen concentration in water from sampling points

Highest concentrations of nitrate-nitrogen were measured in River Cerava inlet (1726.3 mg l^{-1} TP) and River Koselska inlet (670.68 and 667.35 mg l^{-1} TP) during summer and winter. During autumn the lowest concentration has been observed.

Ammonia

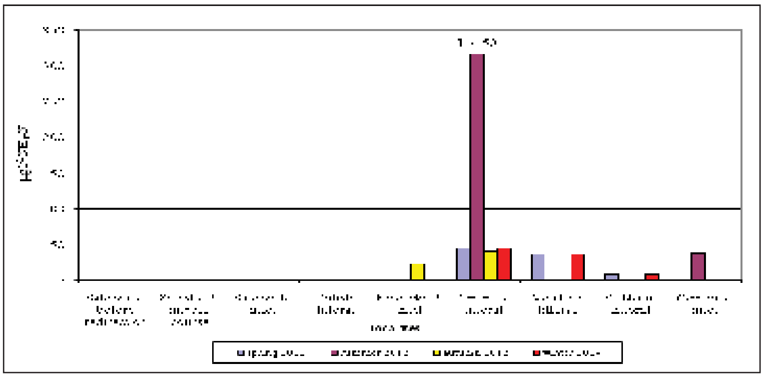

Figure 26: Ammonia - nitrogen concentration in water from sampling points

During all sampling campaigns concentrations have been below LOD (0,001 mg l^{-1}). The highest concentration for ammonia-nitrogen was measured at Grasnica littoral during the summer (316.50 mg l^{-1}).

Total Nitrogen by Kjeldahl

Values for total nitrogen are presented at figure 26.

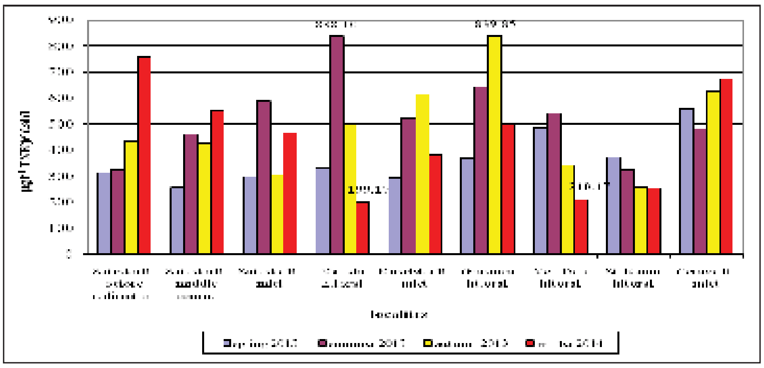

Figure 27: Total nitrogen by Kjeldahl concentration in water from sampling points

Concentrations have been very variable. The highest values were detected at Kallista littoral and Veli dab littoral. The lowest value for total nitrogen by Kjeldahl was measured at 75 m depth during autumn, 172.61mg l^{-1} .

Total nitrogen

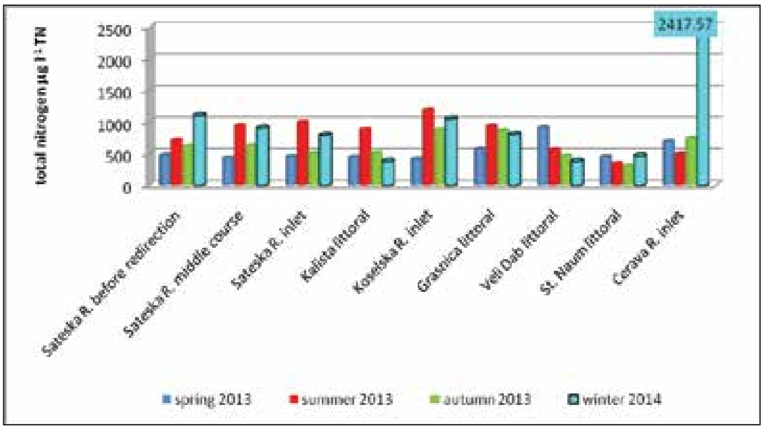

Figure 28: Concentration of total nitrogen in water from sampling points

Highest values for total nitrogen as a sum of organic and unorganic forms of nitrogen has observed in Cerava R. Inlet in winter (2517.57 mg l^{-1} TN). Minimum values for total nitrogen during the winter period are estimated for St. Naum littoral zone (463.0 mg l^{-1} TN), Veli Dab (379.23 mg l^{-1} TN) and Kalista littoral (380.81 mg l^{-1} TN). Generally maximal values for all nitrogen compunds during all sampling campaigns were measured in River Cerava inlet and littoral Grasnica. The lowest valua were measured at Velidab.

According to the OECD classification and Directive for Classification of waters (Official Gazette No 18/99), water quality from St. Naum littoral, Velidab and Kalista belongs to II-III class (oligo-mesotrophic state). Littoral Grasnica is mainly in IV class (meso-eutrophic state). River Sateska before redirection, River Sateska middle course and inlet, River Koselska inlet and River Cerava inlet, according Directive for Classification of waters (Official Gazette No 18/99) belongs from IV-V class (meso-eutrophic state).

Total phosphorus

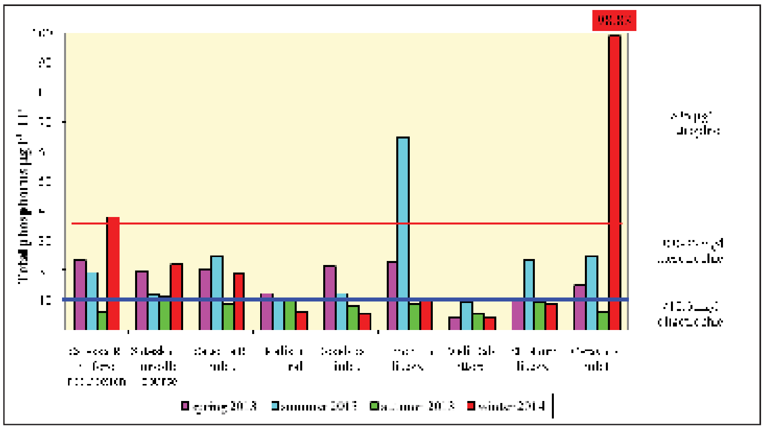

Figure 29: Concentration for total phosphorus in water from sampling points

Highest values for total phosphorus were registered in River Cerava inlet (98.83 mg l^{-1} TP) and River Sateska before redirection (38.14 mg l^{-1} TP) during the winter. During summer the highest value for total phosphorus was registered in the littoral at Grasnica near the river Velgoska mouth (65.01 mg l^{-1} TP).

Generally more values for concentration of total phosphorus during the investigated period belong to mesotrophic state: Sateska River before redirection, Sateska River middle course, Sateska River inlet, Grasnica littoral and Koselska River inlet, (according OECD classification of water), ie IV-V class according Directive for Classification of waters (Official Gazette No 18/99). Sampling sites: Kalista, Koselska R. inlet, Veli Dab and St. Naum belong to oligotrophic state, according OECD classification, ie I-II class according Directive for Classification of waters (Official Gazette No 18/99).   Littoral Grasnica during summer and River Cerava inlet during winter have significantly higher values for total phosphorus. According to the Directive for Classification of waters and OECD belong to IV and V class (eutrophic state).

Trophic state index (TSI)

Carlson's trophic state index is a summary of quick and indicative parameter for presentation of the trophic condition of certain water ecosystems. The calculation of this index is conducted by taking into consideration all characteristics (physical, chemical and biological) of the water represented via suitable researched parameters: Secchi depth (SD), total phosphorus concentration (TP) and chlorophyll *a* concentration (Chla).

Concentration of total phosphorous as a chemical indicator and the transparency as a physical parameter are used as indicators of the trophic condition of the Lakes Ohrid and Prespa. According to the numeric values for the Carlson's Trophic State Index (TSI) based on secchi depth during the investigation, Lake Ohrid belong to **ultra-oligotrophic state**.

According to the numeric values for the Carlson's Trophic State Index (TSI) calculated on basis of the total phosphorus during the investigated period Lake Ohrid is in stable oligotrophic state.

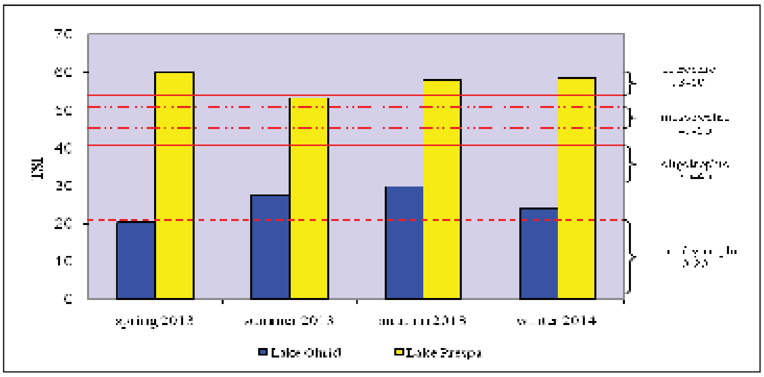

Figure 30: Trophic state index (TSI) for Lakes Ohrid and Prespa

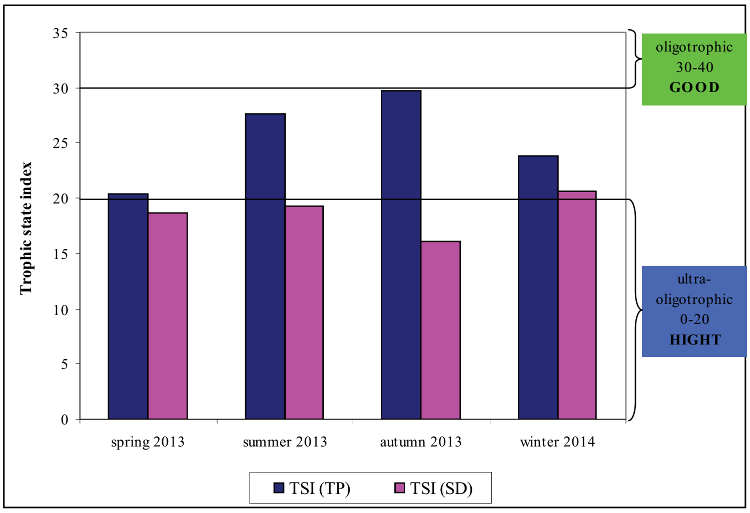

Figure 31: Trophic state index (TSI) for Lake Ohrid littoral zone

The values of the Trophic state index (TSI) at the littoral zone of Lake Ohrid based on the concentration of total phosphorus indicate a great seasonal variability. According to this littoral zone of Lake Ohrid (Kalista, Veli Dab and St. Naum) belong mainly to the **oligotrophic state**. Exception from this condition is littoral Grasnica where the values for TSI during the spring and summer period belonged to the **mesotrophic state**. During summer value for TSI for St. Naum has been in mesotrophic state too.

The values of the Trophic state index (TSI), at the littoral zone of Lake Ohrid, based on concentration of total phosphorus indicate to a great seasonal variability .

Sediment analyses

The organochlorine pesticides measured in sediments from St. Naum, River Sateska, River Koselska and Grasnica are: *gamma*-HCH (□-HCH), □HCH (sum of □-isomer, □-isomer and □-isomer), endosulfan (total of a and b endosulfan), DDT metabolites (*p,p*'–DDE, *p,p*'–DDD and *p,p*'-DDT). The most abundant of the detected organochlorine pesticides was the sum of DDT metabolic forms, i.e. *p,p*'-DDT, *p,p*'-DDE and *p,p*'-DDD. For sediment sample collected from St Naum, value for content of gamma-HCH is below LOD, for Sum HCH 0.42 µg kg^{-1} dry sediment, for endosulfan 0.491 µg kg^{-1} dry sediment and for total DDT 1.389 µg kg^{-1} dry sediment. Dominant metabolic form of DDT in the sediment from St. Naum was p,p'-DDE (0.668 µg kg^{-1} dry sediment).

For sediment samples collected from Grasnica values for organochlorine pesticide are higher than at St. Naum. The value for content of gamma-HCH is below 0.291 µg kg^{-1} dry sediment, for total HCH 1.018 µg kg^{-1} dry sediment, for endosulfan 0.849 µg kg^{-1} dry sediment and for total DDT 2.435 µg kg^{-1} dry sediment. Dominant metabolic form of DDT in the sediment from littoral Grasnica was p,p'-DDE (1.721 µg kg^{-1} dry sediment)

The existence of organochlorine pesticides in the sediment is mainly due to the chemical stability of these compounds, their high lipid solubility and the bioaccumulation of this group of persistent organic pollutants (POP's) rather than their current use. The detected concentrations are clearly below toxic thresholds and consequently severe effects on the endemic species of Lake Ohrid are not very likely but still not completely understood.

6. 3. Classification

- Carlson's trophic state index is summary, quick and indicative parameter for presentation of the trophic condition of certain water ecosystems. The calculation of this index is conducted by taking into consideration all characteristics (physical, chemical and biological) of the water represented via suitable researched parameters: Secchi depth (SD), total phosphorus concentration (TP) and chlorophyll a concentration (Chlaa).

- Concentration of total phosphorous as a chemical indicator and the transparency as a physical parameter are used as indicators of the trophic condition of the Lake Ohrid. According to the numeric values for the Carlson's Trophic State Index (TSI), based on secchi depth during the investigation Lake Ohrid belong to ultra-oligotrophic state.

- According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus during the investigated period Lake Ohrid is in stable oligotrophic state. According OECD clasification and Directive for Classification of Water Lake Ohrid belong to oligotrophic state, ie I class of water.

- According OECD classification and Directive for Classification of waters (Official Gazette No 18/99), based on concentration of total nitrogen the water quality from St. Naum littoral, Velidab and Kalista belongs to II-III class (oligo-mesotrophic state). Littoral Grasnica is mainly in IV class (meso-eutrophic state). River Sateska before redirection, River Sateska middle course and inlet, River Koselska inlet and River Cerava inlet, according Directive for Classification of waters (Official Gazette No 18/99) belongs from IV-V class, ie meso-eutrophic character.

- Generally more values for concentration of total phosphorus during the investigated period belong to mesotrophic state: Sateska River before redirection, Sateska River middle course, Sateska River inlet, Grasnica littoral and Koselska River inlet, (according OECD classification of water), ie IV-V class according Directive for Classification of waters (Official Gazette No 18/99). Sampling sites: Kalista, Koselska R. inlet, Veli Dab and St. Naum belong to oligotrophic state, according OECD classification, ie I-II class acording Directive for Classification of waters (Official Gazette No 18/99). Littoral Grasnica during summer period, and River Cerava inlet during winter have significantly higher concentration for total phosphorus and according Directive for Classification of waters and OECD clasification belong to IV and V class, ie eutrophic state.

- The values of the Trophic state index (TSI), at the littoral zone of Lake Ohrid, based on concentration of total phosphorus indicate to a great seasonal variability. According this index, littoral zone of Lake Ohrid (Kalista, Veli Dab and St. Naum), belong mainly to the oligotrophic state. Exception from this condition is littoral Grasnica where the values for TSI during the spring and summer period belong to the mesotrophic state. During the summer period value for TSI for St. Naum is in mesotrophic state too.

7. Recommendations

The worst conditions (according to all analyzed indicators) were observed in Grasnica where the water is mainly mesotrophic and sometimes transferring into eutrophic character. River Velgoska (which is nearby) has been suggested as the main cause, since it is the end-recipient of waste, drainage and household waters.

During the summer time (touristic season) high values for nutrients concentrations were registered in the analysed localities (concentrations for total phosphorus and total nitrogen are with maximum values). The organic loading is more intense during the summer period too. This condition occurs as a result of the increased number of people in that period and the increased number of touristic facilities, both of which contribute to the pollution of the ecosystem.

Because of that it is important to have continued monthly analyses of the water quality based on the physico-chemical parameters in the littoral and pelagic zone of the Lake Ohrid and tributaries which gravitate towards this ecosystem. In the future the number of samples along the rivers (from source to input to the lake) should be increased including the measurement of the water velocity of the rivers for the determination (calculation) of the nutrient input from the rivers (nutrient and organic balances).

8. Summary

From the investigations are evidenced increased nutrient concentrations in the littoral zone (especially near the rivers mouth), of Lake Ohrid during the summer period. Lake Ohrid is a very attractive tourist destination (especially in the summer) On the other hand this is the result from the high influence of the tributaries in the watershed of the lake. The affected tributaries are discharged system for the communal and industrial waters as well as for the drainage waters from the surrounding agricultural area. The water from rivers with this quality entirely migrates to the littoral zone which is to be considered as their end recipient and prime risk for this aquatic ecosystem. Phosphorus released in the littoral zone could be transported to the deep pelagic lake water as a result of the convective night movement and as a result of the lake's mainstream.

The most alarming condition for all investigated period has been noticed in the Cerava R. Inlet where a tendency to an eutrophic state could be observed. TSI for this point is 70.42. From the obtained results, River Cerava is a strongly polluted river which low water flow. It flows through two countries, Albania and Macedonia (flows through mining region Albania and through distinctly agricultural region Macedonia).

According to OECD classification and Directive for Classification of waters (Official Gazette of R.M. No 18/99), based on concentration of total nitrogen, the water quality from St. Naum littoral, Velidab and Kalista belongs to II-III class (oligo-mesotrophic state). Littoral Grasnica is mainly in IV class (meso-eutrophic state). River Sateska before redirection, River Sateska middle course and inlet, River Koselska inlet and River Cerava inlet, according to the Directive for Classification of waters (Official Gazette of R.M. No 18/99) belongs from IV-V class, ie meso-eutrophic state.

According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus Lake Ohrid has still an oligotrophic state. According to OECD classification and Directive for Classification of Water Lake Ohrid belong to oligotrophic state, I class of water.

The values of the Trophic state index (TSI), at the littoral zone of Lake Ohrid, based on concentration of total phosphorus indicate to a great seasonal variability. According to this index littoral zone of Lake Ohrid (Kalista, Veli Dab and St. Naum) belong mainly to the oligotrophic state. Exception from this condition is littoral Grasnica where the values for TSI during the spring and summer period belong to the mesotrophic state. During the summer period value for TSI for St. Naum is in mesotrophic state too.

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UNIVERSITETI I TIRANËS
FAKULTETI I SHKENCAVE TË NATYRËS
DEPARTAMENTI I BIOTEKNOLOGJISË
Adresa: Bulevardi " Zogu I ", Nr. 25/1, Tiranë, Tel. & Fax: +355 4 2229590. www.fshn.edu.al

COMPREHENSIVE REPORT ON PHYTOPLANKTON ANALYSIS BASED ON PIGMENT CONTENT AND TENTATIVE ASSESSMENTS OF WATER QUALITY

SAMPLING CAMPAIGNS OF 2013-2014 FOR LAKES OHRID AND PRESPA

1. Contents

Based on the general terms of the contracts between the Deutsche Gesellschaft für International Zusammenarbeit (GIZ) of the German Development Cooperation, Project CSBL and Prof. Asoc. Dr. Ariola Bacu, the main objectives of the following report were to conduct phytoplankton analysis based on chlorophyll *a* content, the tentative assessment of water quality based on the index of Carlson (TSIC), and the phytoplankton analysis based on chemotaxonomy at Lake Prespa for the campaign of April 2014.

1. Stations of sampling

Chlorophyll a as an indirect indicator of phytoplankton biomass was measured at two stations of Lake Prespa and three stations at Lake Ohrid, respectively:

1. Lake Prespa, Station of Gollomboc, 100 m from the shore
2. Lake Prespa, Station of Pustec, 100 m from the shore

1. Lake Ohrid, Station of Lin, 200-250 m from the shore / 3,5 m depth
2. Lake Ohrid, Station Memlisht/Gur i Kuq, 200-250 m from the shore / 6 m depth
3. Lake Ohrid, Station Pogradec/Tushemisht, 200 m from the shore / 5,5 m depth

This phytoplankton indicator was measured during four campaigns, respectively in July-October 2013 and February 2014 for Lake Ohrid, and July-October 2013 / February-April 2014 for Lake Prespa.

2. Methods followed for chlorophyll a measurement and trophic state evaluation

Filtering of water and extraction of chlorophyll *a* for the Albanian stations at Lakes Ohrid and Prespa was conducted as follows:

Sampling of the water from the appointed stations was conducted in polyethylene bottles of about 2 liters. They were kept in cold dark boxes until filtered within three hours.

Filtration, extraction and determination of the concentration of chlorophyll *a*:

- The reported volume of water was filtered in a Vacuum Water Filtering System on a filter GF/F (0.7 μm); the pump pressure did not exceeded 25 Kpa (about 150 mm Hg)
- The filter is transferred in a test tube and covered with 90% acetone.
- The filter was kept overnight in the dark at 4°C for pigment extraction.
- The filter was cut into pieces and homogenized in 90% acetone for 2 minutes in a glass mortar.
- The extract was collected in centrifuge tubes.
- Tubes were vortexed.
- Tubes were filled with acetone up to the reported volume, equal for all the samples.
- The extract was centrifuged 15 minutes/ 4000 rpm.
- The supernatant was collected in a cylinder and the volume was measured.
- The supernatant was used to fill the spectrophotometric cuvette and the optical density was measured at 664 and 750 nm, using as blank another cuvette filled with acetone 90%.
- The concentration of chlorophyll *a* was calculated based on the formula:

Chlorophyll *a* (µg/dm^3) ={[A(s,664,i)- A(b,664,i)]-[A(s,750,i)-A(b,750,i)].v.106/ (□.CO. V)

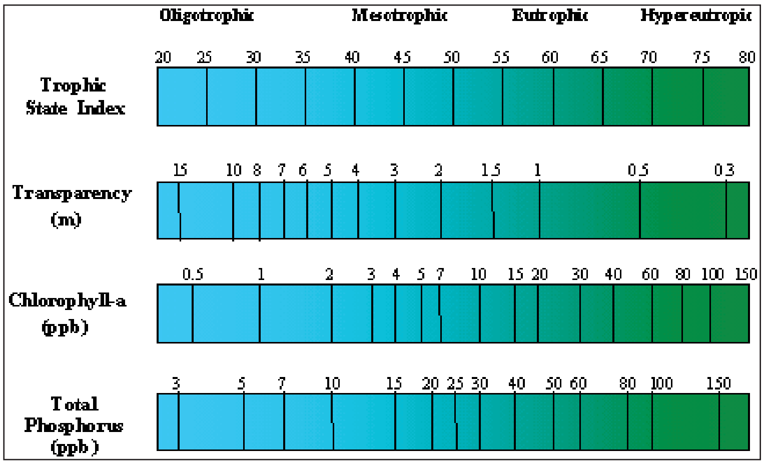
1. A(b,664,i)= absorbance of blank in 664 nm
2. A(b,750,i)= absorbance of blank in 750 nm
3. A(s,664,i)= Absorbance of blank in 664 nm
4. A(s,750,i)= absorbance of blank in 750 nm
5. Є= coefficient of absorption of chlorophyll in acetone 90% (87,67 dm^3/cm.g)
6. CO= optical length of cuvvete (cm)
7. v = volume of extract (cm^3)
8. V= volume of filtered sample (cm^3)

2.1. Classification of the trophic state

The Index of Trophic State (TSI) according to Carlson, 1977 served to characterize the trophic state at sampling stations of Lake Prespa at the Albanian side. Values of TSI based on chlorophyll *a* content were calculated as follows:

TSI for chlorophyll *a* (TSIC) = 9.81*[ln(chlorophyll-*a*)] + 30.6

The scale of TSI (according to Carlson 1977, below) varies from 0 (ultra-oligotrophic) up to 100 (hyper-eutrophic). Chlorophyll *a* were measured in microgram/litre (mg/l).



2.2. Pigment analysis (for Lake Prespa during the campaign of April 2014)

Together with chlorophyll *a*, chlorophyll *b*, chlorophyll *c1+c2*, and carotenoids were measured using the spectrophotometric acetone trichromatic method and equations based on absorption maxima for each component respectively (with coefficients of Jeffrey and Humphrey, 1975). The absorbances allow for quantitative evaluations of the chlorophyll categories present at the stations (chlorophyll *a* is measured at 664 nm, chlorophyll *b* at 647 nm, chlorophyll *c* at 630 nm, and carotenoids at 480 nm and 510 nm). For samples taken at the Albanian side of Lake Prespa, the spectrophotometric measurement of photosynthetic pigments was used to describe phytoplankton composition.

3. Results For Lake Ohrid

Measurement of *chlorophyll a* content during three campaigns of July and October 2013, and February 2014 gave the following results for the three Albanian stations at Lake Ohrid.

Lake Ohrid - Station 1	July 2013	Station 1	1.910 µg/dm³
Lake Ohrid - Station 2	July 2013	Station 2	0.467 µg/dm³
Lake Ohrid - Station 3	July 2013	Station 3	0.478 µg/dm³
Lake Ohrid - Station 1	October 2013	Station 1	0,920 µg/dm³
Lake Ohrid - Station 2	October 2013	Station 2	0,699 µg/dm³
Lake Ohrid - Station 3	October 2013	Station 3	0,643 µg/dm³
Lake Ohrid - Station 1	February 2014	Station 1	0,594 µg/dm³
Lake Ohrid - Station 2	February 2014	Station 2	0,643 µg/dm³
Lake Ohrid - Station 3	February 2014	Station 3	0,923 µg/dm³

The trophic state, according to the above-cited methodology, for the three stations at the Albanian side of Lake Ohrid is oligotrophic (Annex 1).

4. Results for Lake Prespa

Chlorophyll *a* content measured during the July-October 2013 and February-April 2014 sampling campaigns at the Albanian side of Lake Prespa were as follows.

July 2013		
Lake Prespa	Station Gollomboc	2.290 µg/dm³
Lake Prespa	Station Pustec	3.226 µg/dm³
October 2013		
Lake Prespa	Station Gollomboc	5.116 µg/dm³
Lake Prespa	Station Pustec	5.811 µg/dm³
February 2014		
Lake Prespa	Station Gollomboc	2.952 µg/dm³
Lake Prespa	Station Pustec	2.874 µg/dm³
April 2014		
Lake Prespa	Station Gollomboc	1.412 µg/dm³
Lake Prespa	Station Pustec	1.633 µg/dm³

Trophic State (Lake Prespa). Values of TSIC calculated according to Carlson (1977) characterize both stations as mesotrophic throughout the year except during the sampling of April 2014, which was characterised by a low Chl. *a* concentration corresponding to an oligotrophic state. The decline in Chl. *a* is common for Prespa Lake during the early season of the year (from March to May the phytoplankton biomass is at its minimum, it later increases until October), but values generally remain within the mesotrophic state (Annex 1).

Chemotaxonomy (Lake Prespa). For the purpose of *chemotaxonomical determination* of phytoplankton assemblages at Lake Prespa in April 2014, the following pigments were measured spectrophotometrically: chlorophyll *a*, chlorophyll *b*, chlorophyll *c1+c2*, and carotenoids, yielding the following results.

April 2014		Chlb	Chlc1+c2	Carotenoids
Lake Prespa	Station Gorice	0.457 μg/dm³	0.757 μg/dm³	2.826 μg/dm³
Lake Prespa	Station Pustec	0.502 μg/dm³	0.867 μg/dm³	3.204 μg/dm³

For the analysis of the taxonomic composition of phytoplankton at stations on the Albanian side of Lake Prespa, an alternative method was used to analyse algal pigments. Chlorophyllian pigments have many favourable characteristics as chemotaxonomic markers. They are present in all photosynthetic algae but not in most bacteria, protozoa or detritus, allowing phytoplankton to be distinguished from other components of the microbial community. Many pigments are limited to particular classes or even genera, allowing the taxonomic composition of the phytoplankton to be determined to class level or better. They are strongly colored, and in the case of chlorophylls and phycobiliproteins, fluorescent at visible wavelengths, allowing them to be sensitively detected. They are labile and rapidly degraded after the death of the cell, thus distinguishing live from dead cells (S.Wright, 2005). Comprehensive data and graphics sheets are compiled for 47 of the most important chlorophylls and carotenoids in marine algae (Jeffrey *et al., 1997).* HPLC and spectrophotometric or fluorometric analysis of Chl. *a* are the most appropriate techniques to measure pigment concentrations. The accuracy of spectrophotometry, fluorometry, and HPLC were compared in the SCOR/UNESCO volume (Mantoura *et al*, 1997). The absorbances of samples measured at five wavelengths serve for the qualitative evaluation of the three chlorophyll categories, chlorophyll *a* measured at 664 nm, chlorophyll *b* at 647 nm, chlorophyll *c* at 630 nm, and carotenoids at 480 nm and 510 nm. CHEMTAX software was developed in cooperation with CSIRO to determine the contribution of major algal components to total chlorophyll *a (Mackey et al. 1996,* Wright *et al. 1996*). It is based on matrix factorization.

According to a number of reports (S.W.Wright 2005; Ston J. & Kosakowska A., 2000; USEPA Method 446.0, 1997, etc.), the presence of certain pigments corresponds to specific classes of phytoplankton, and so does the ratio among different pigments.

Table 1 shows the results of the chemotaxonomic analysis of water samples collected at two sampling stations at the Albanian side, taking into consideration the presence of different classes of pigments, which belong to different phytoplankton classes, and ratios among pigments to determine which predominant classes among those possibly present (Table 2)

Station	Chl a	Carotenoids / Chl a	Chl b / Chl a	Chl c / Chl a	
Gollomboc, April 2014	1.412	2	0.323	0.536	*Chlorophyta (included Prasinophyta); Diatoms A- B (Bacillariophyta); Dinoflagelates; Euglenophyta, Cyanophyta, Rapidophyta, Haptophyta, Chrysophyta.*
Pustec, April 2014	1.633	1.96	0.307	0.530	

Table 1. Predicted phytoplankton composition based on the presence of different classes of pigments. (The taxonomy used acc. to Not *et al*, 2012; Tirjatkin, 2011 etc.).

Station	Chl. a	Chl. b	Chl. c (c1+c2)	Carotenoids	Phytoplankton Composition
Gorricë, April 2014	1.412	0.457	0.757	2.826	*Prochlorophyta, Chlorophyta (Prasinophyta), Bacillariophyta (Diatoms), Cyanophyta, Euglenophyta, Chrysophyta, Rapidophyta, Haptophyta, Dinophyta, Cryptophyta, Bolidophyta*
Pustec, April 2014	1.633	0.502	0.867	3.204	*Prochlorophyta, Chlorophyta (Prasinophyta), Bacillariophyta (Diatoms), Cyanophyta, Euglenophyta, Chrysophyta, Rapidophyta, Haptophyta, Dinophyta, Cryptophyta, Bolidophyta,*

Table 2. Phytoplankton classes based on the ratios among pigments.

4.1. Comparison of the results of classical taxonomy and Chemotaxonomy

During the sampling campaign of April 2014 for Lake Prespa the categories of phytoplankton were determined according to chemotaxonomy and classical methods. Based on both methods:

The categories of phytoplankton present at both stations of Lake Prespa are the same. The divisions of phytoplankton identified based on the classical method are Bacillariophyceae, Chlorophyceae, Dinophyceae, Cyanophyceae and Chrysophyceae.

The main divisions of phytoplankton identified based on chemotaxonomy were diatoms (Bacillariophyceae); Chlorophyta (including Prasinophyta), Dinoflagellates, Cyanobacteria (Cyanophyceae), Chrysophyta (Chrysophyceae), Euglenophyta, Haptophyta, Rapidophyta. (The taxonomy used acc. to Not *et al*, 2012; Tirjatkin, 2011).

Based on the classical method, a number of species were identified in each division while based on chemotaxonomy, the classification goes up to class level.

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6. Annexes: Summary of results

Annex 1. Chlorophyll *a* concentrations and TSI (Chl. *a*) at Lake Ohrid (Albania) from April 2013 to February 2014

		Sampling point					
		Lin		Memlisht/Gur i Kuq		Pogradec/Tushemisht	
Month	**Parameter**	Chlorophyll a (µg/L)	Trophic State Index (TSI) [1]	Chlorophyll a (µg/L) [1]	Trophic State Index (TSI)	Chlorophyll a (µg/L)	Trophic State Index (TSI) [1]
July 2013		1.90	37.0	0.47	23.0	0.48	23,4
October 2013		0.92	30.0	0.70	27.0	0.64	26
February 2014		0.59	25.5	0.64	26.3	0.92	29.8
Overall assessment of trophic state		oligotrophic		oligotrophic		oligotrophic	

[1] According to Carlson (1977)

Annex 2. Chlorophyll *a* concentrations and TSI (Chl. *a*) at Lake Ohrid (Albania) from April 2013 to February 2014 and assessment of the composition of phytoplankton assemblages based on quantitative morphological versus qualitative chemotaxonomic analyses (April 2014 only)

		Sampling point							
		Gorice / Gollomboc				Pustec			
Month	**Parameter**	Chlorophyll a (µg/L)	Trophic State Index (TSI) [1]	Three most abundant groups by method (April 2014 only): morphological (cells/L)	chemotaxonomic (qualitative)	Chlorophyll a (µg/L)	Trophic State Index (TSI) [1]	Three most abundant groups by method (April 2014 only): morphological (cells/L)	chemotaxonomic (qualitative)
July 2013		2.29	38.7			3.23	42.1		
October 2013		5.12	47.0			5.81	48.0		
February 2014		2.95	41,2			2.87	41.0		
April 2014		1.41	34.0			1.63	35.4		
- Cyanophyta					x				x
- Bacillariophyta				12.8×10^3	x			15.0×10^3	x
- Chlorophyta					x				x
- Chrysophyta				3.2×10^3				3.3×10^3	
- Pyrrophyta (Dinophyta)				6.7×10^3				7.8×10^3	
- Euglenophyta									
Overall assessment of trophic state		mesotrophic				mesotrophic			

[1] According to Carlson (1977)

Chlorophyll a of Lake Ohrid

–Final Report –

Dr. Suzana Patceva
Hydrobiological Institute - Ohrid

June, 2014

Contents

1. Introduction

Phytoplankton biomass is usually measured by the amount of chlorophyll *a* in the water.

Chlorophyll *a* is a photosynthetic pigment that serves as a measurable parameter for all phytoplankton production. On average, 1.5% of algal organic matter is chlorophyll a (Raschke, 1993). Thus, if chlorophyll *a* concentrations are known, a manager can estimate the phytoplankton biomass in the water body (Nürnberg & Shaw, 1999; Raschke, 1993; Marshall and Peters, 1989). High biomass will discolour the water body.

Chlorophyll *a* concentration and Secchi depth are both the most significant measures of the lake trophic state (Carlson, 1977). Chlorophyll *a* represents the biological response of the lake, all other factor being equal.

2. Methods

Sampling points

Investigations of chlorophyll a concentration in Macedonian part of Lake Ohrid comprised sampling from 1 sampling point in the pelagic zone of Lake Ohrid on 9 depths of the water column.

Sampling depths (9):

- 0.5m
- 10m
- 20m
- 30m
- 40m
- 50m
- 75m
- 100m
- 150m

Figure 1. Sampling point for chlorophyll a investigation in Lake Ohrid

Sampling campaigns

Phytoplankton samples were taken during two sampling campaigns: first in spring period and the second in summer period.

The weather in the first campaign was sunny and the temperature of surface water was 10˚C.

In the second campaign the weather was sunny and the temperature of surface water was 23.2˚C.

Sampling and analyses

1. For the chlorophyll a concentration, samples were taken with the Niskin water sampler. Samples in 1 l polyethylene bottles were transport to the laboratory in hand refrigerators.
2. Chlorophyll a analyses were made with Spectrophotometer UV-VIS SPECORD 10 (Zeiss) after extraction with 90% ethanol according to ISO 10260 (1992).
3. The calculation of trophic state index (TSI) was done according to Carlson (1977).

3. Existing data and gaps

More investigations for the phytoplankton and chlorophyll *a* as indicators of trophic state in the littoral zone of Lake Ohrid and influence of the tributaries on the trophic state and on the phytoplankton composition and production were carried out (Mitic, 1985, 1993; Mitic , V. & Patceva, S., 1999; Patceva, S. & V. Mitić, 2001; Patceva, 2001; Mitić, V. & S. Patceva, 2002; Patceva, S. & V. Mitić, 2002; Patceva et al. 2003; Patceva, S. & V. Mitić, 2003; Patceva et al. 2004 a,b; Patceva, 2005; Patceva et al. 2005; Patceva et al. 2007) and the results from all of these investigations indicated that region Grasnica, where inflows River Velgoska had the worst trophic state and this region introduce large danger for the trophic state of Lake Ohrid in whole. Afterwards respectfully follows Cerava, Sateska and Koselska.

In the last few decades, as is known, the increased anthropogenic impact has negative effect in the littoral region, but slowly this impact is spreading in the pelagic zone of the oligotrophic Lake Ohrid, too. The last investigations of the phytoplankton in the pelagic zone of Lake Ohrid were carried out within the MSE projects "Biodiversity and ecology of plankton communities in Lake Ohrid (Macedonia) and Plitvice Lakes (Croatia)" (2007-2009) and "Spatial and temporal changes in planktonic community –carrier of the Lake Ohrid trophic state (2009-2011)". Subject of the investigations in the projects were pelagic zone of the Lake Ohrid and the complex investigations of the phytoplankton were done, as well as the content of chlorophyll *a*, as important parameter for the water trophic state. According to the obtained values from the phytoplankton investigations, Lake Ohrid is in stable oligotrophic condition without notably signs of eutrophication, with the exception of minor changes in dominant species of algae. Algal production was not increased in comparison with past investigations and the tendency of mildly decreasing was noted, especially during the summer period.

Because phytoplankton is the most sensitive indicator of the water trophic state, especially in the pelagic zone of the lakes, continued investigation is needed, but from 2011 to today are missing investigations as in pelagic and in the littoral zone of Lake Ohrid. It is important because in recent years significant climate changes and increased anthropogenic pressure were occurred.

4. Results

The seasonal distribution of chlorophyll *a* concentration in Lake Ohrid was opposite than in Lake Prespa. Chlorophyll *a* concentration in Lake Ohrid in the water layer between 0m and 30m depth reached higher values in the spring period and significantly decreased in the summer period (Fig. 2). Especially low values were registered in the surface layer.

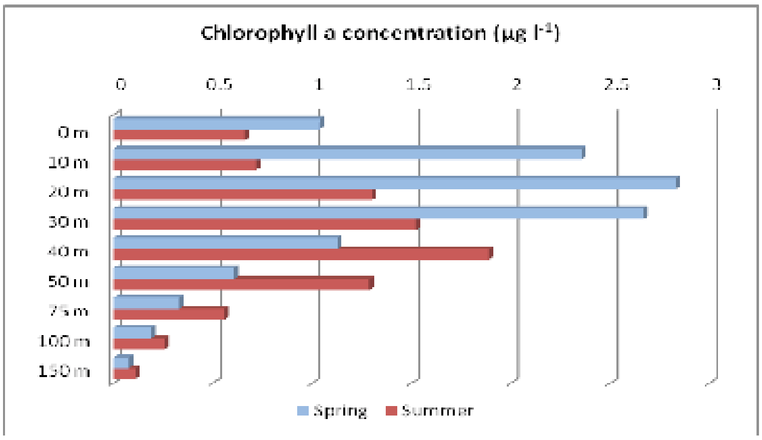

Figure 2. Chlorophyll *a* concentration in the pelagic zone of Lake Ohrid

In the water layer from 40m to 150m depth higher values were observed in the summer period.

Values of chlorophyll *a* concentration in Lake Ohrid varied between 0.08 mg l^{-1} at 150 m depth and 2.83 mg l^{-1} at 20 m depth (Fig. 2).

The highest average values of chlorophyll *a* concentration in the water layer of Lake Ohrid were observed in the layer between 20 and 40 m depth where phytoplankton abundance was the highest (Fig. 3).

This kind of chlorophyll *a* distribution in Lake Ohrid in full coincide with the general distribution of chlorophyll *a* in the oligotrophic lakes from temperate zone, obtained from Marshall and Peters, (1989).

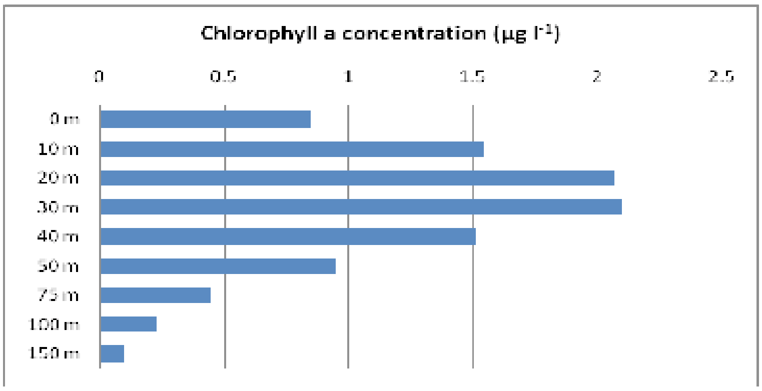

Figure 3. Average chlorophyll *a* concentration (µg l^{-1}) in Lake Ohrid

Trophic state index (TSI; Carlson, 1977) is measurement of the water trophic level. It provides the basis that ties chlorophyll *a* levels with total phosphorus, which tends to fuel algal productivity. Calculation of this index is based on chlorophyll *a* concentration (Chl) and this concentration is indirect measure of phytoplankton biomass:

TSI value

- Trophic level
- < 40
- Oligotrophy
- 40 - 50
- Mesotrophy
- 50 - 60
- Eutrophy I
- 60 - 80
- Eutrophy II
- > 80A
- Hypertrophy

Table 1. Trophic state index (TSI) during two sampling campaigns in Lake Ohrid

	Pelagial 0m
Spring	30,95
Summer	26,49

Table 2. Trophic state categories of lakes predicated on summer chlorophyll a concentration based on Nürnberg (1996)

Oligotrophy	Mesotrophy	Eutrophy	Hypereutrophy
< 3,5	3,5 - 9	9,1 -25	> 25

Table 3. Trophic state of Lake Ohrid during summer sampling campaigns based on Nürnberg (1996)

	Pelagial 0m
Summer	0,66

According to the trophic state index (TSI) Lake Ohrid is in oligotrophic state (Tab. 1).

According to the classification system of Nürnberg (1996) which are regard to summer period, Lake Ohrid is in oligotrophic state (Tab. 3).

In accord with Water Framework Directive we can define status of pelagic zone of Lake Ohrid as **high** (Tab. 4).

Table 4. Ecological Status of Lake Ohrid according to WFD

Spring	
Summer	

5. Conclusions

According to the trophic state index (TSI) obtained during these investigations, Lake Ohrid is in oligotrophic state.

According to the classification system of Nürnberg (1996) which are regard to summer period, Lake Ohrid is in oligotrophic state, too.

The pelagic zone remains immune to the anthropogenic pressures that threatened these waters in previous decades.

During the first (spring) sampling campaign chlorophyll *a* concentrations in the water layer between 0 and 30 m depth were significantly higher than in the second (summer) sampling campaign.

The highest average values of chlorophyll *a* concentration in the water layer of Lake Ohrid were observed in the layer between 20 and 40 m depth

6. Recommendations

In accord with Water Framework Directive we can define status of pelagic zone of Lake Ohrid as high.

The seasonal pattern of phytoplankton biomass in temperate lakes is frequently described as a pronounced spring bloom followed by a summer depression, a subsequent fall bloom, and low levels through winter (Goldman and Horne 1983; Taub 1984).

Considering that Lake Ohrid is a large deep lake, with specific spatial and temporal phytoplankton distribution, it is recommended a monthly or seasonal frequency of sampling of phytoplankton and chlorophyll *a* parameters with sampling points in the pelagic and littoral zone, especially in the areas which are under anthropogenic pressure.

Pelagic zone of Lake Ohrid with sampling points at nine different depths is strongly recommended to be sampling point in the future monitoring and it should be considered as reference.

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FINAL REPORT BY THE DEPARTMENT OF HYDROBOTANY

Hydrobiological Institute – Ohrid

R. Macedonia

Prepared by:

d-r Marina Talevska

d-r Sonja Trajanovska

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Contents

1. INTRODUCTION

Aquatic macrophytes which are found in the littoral zones of lakes comprise macroscopic forms of vascular plants, a small number of aquatic mosses, and some large algae such as *Chara* and *Nitella* species, as well as *Cladophora* – (Wetzel, 1975; Lind, 1979). They are relatively long-lived organisms (their life span is measured in months or years) and are often used as key indicators of the ecological status of lakes (Solimini et al., 2006).

Aquatic macrophytes in the littoral zones of lakes have two fundamental properties which make them useful and attractive as limnological indicators: 1) they react slowly and progressively to changes in nutrient conditions (Melzer, 1999); 2) the littoral zone may experience patterns of nutrient (and pollutant) concentrations caused by natural or artificial inflows as well as by diffuse, non-point sources (Dave, 1992; Drake & Heaney, 1987 in Melzer, 1999) and rooted submerged macrophytes may reflect this patchiness. Consequently, macrophyte indices reflecting nutrient pollution have been developed in Europe for both lakes (Melzer, 1999) and rivers (Schneider et al. 2000; Schneider & Melzer 2003).

Macrophytes are one of the basic elements used in addition to phytoplankton, macrozoobenthos and fish in the assessment of the ecological status and classification of waters.

The monitoring of all biological elements, including macrophytes, requires the development of two complementary methodologies:

- **Field survey, i.e. the means of material sampling;**
- **Assessment of water status based on the material collected in the field, i.e. the means of data analysis and calculation of biological indicators.**

2. LAKE OHRID

Lake Ohrid is the largest and the deepest lake in the Dassaret lake group. It fills the deepest part of the Ohrid valley. At a sea level of 693 m, Lake Ohrid has a surface area of 358 km^2, a maximum length of 30.5 km and a maximum width of 15 km. Lake Ohrid has a maximum depth of 289 m, and a great transparency of the water (21 meters). Moreover, it is supplied with water from surface springs, sublacustric springs and tributaries (Rivers Koselska, Velgoska, Sateska and Cherava). The River Crn Drim outflows from the Lake.

2.1. Monitoring of macrophytes in tributaries of Lake Ohrid

For the monitoring of macrophytes at the tributaries of Lake Ohrid – River Sateska before redirection, River Sateska middle course, River Sateska inlet, River Koselska inlet and River Cherava inlet (Fig.1.) – the following analyses were conducted :

- **Collection of plant material during the period of maximum growth (middle of summer period - July and August),**
- **Preparation of the list of species presented in the analysed sites (qualitative composition of macrophytes),**
- **Estimation of plant abundance of presented species in the analysed sites (quantitative composition of macrophytes),**
- **Determination of collected materials,**
- **Analysis of the trophic state of the analysed sites according to the registered macrophytes.**

Figure 1. Map of Lake Ohrid with sampling sites - tributaries of Lake (green)

2.2. The methodology of the field survey

The plant material was collected during the period of maximum growth (middle of summer period – July and August), using the methods of Wetzel & Likens 1979 and Schneider & Melzer 2003. In shallow water, in which it is possible to collect the samples by hand, entire plants were collected. In deeper water, the plants were collected using a rake. The plant material was washed carefully to remove sediments, epiphytes and animals.

All species difficult to identify in the field were stored in plastic bags for further analyses; bigger plants were dried on a newspaper; small, thin leaved or fragile plants were preserved in alcohol.

Furthermore, plant abundance of the species present at the researched sites was determined (quantitative composition of macrophytes) according to a five-point scale (Tüxen & Preising, 1942 in Melzer, 1999), whereby 1 = very rare, 2 = infrequent, 3 = common, 4 = frequent, 5 = predominant (abundant).

Figure 2. Sample collection at the tributaries of Lake Ohrid

2.3. Laboratory work

Collected material was determined in a laboratory using different floras and keys: for vascular macrophytes: Hayek, ed. (1924-1933); Josifović, ed. (1970-1977); Tutin et al., ed. (1964-1980), and for charophytes: Corillion (1957, 1975), Golerbah & Krasavina (1983), Krause (1997), Schubert & Blindow (2003) and Wood & Imahori (1964, 1965).

Additionally, a list of species found at each sampling site was compiled (qualitative composition of macrophytes) according to the analysed macrophytes and the trophic state of the researched sites. Accordingly, a catalogue of nine macrophyte indicator groups was used, their scores ranging from 1 to 5 (Melzer & Schneider, 2001), where each group of macrophytes exhibits different sensitivity towards nutrient enrichment.

2.4. Results and Discusion

Table 1. Composition and abundance of the macrophytes (vascular macrophytes and charophytes) at the tributaries of Lake Ohrid.

TRIBUTARIES OF LAKE OHRID						
		Sateska (before redirection)	Sateska (middle course)	Sateska (inlet)	Koselska (inlet)	Cherava (inlet)
1.	*Phragmites australis*					2
2.	*Typha latifolia*		2			1
3.	*Typha angustifolia*				2	1
4.	*Petasites hibridus*				3	
5.	*Polygonum hidropiper*	2	2			
6.	*Mentha longifolia*		2			
7.	*Epilobium hirsutum*	4	3			
8.	*Rumex palustris*		2		1	
9.	*Sparganium erectum*				2	
10.	*Fontinalis antipyretica*			2	2	
11.	*Ranunculus circinatus*				2	
12.	*Elodea canadensis*		1			
13.	*Lemna minor*		1			
14.	*Potamogeton perfoliatus*			1		
15.	*Berula erecta*		2		1	
16.	*Calitriche verna*		2		1	
17.	*Roripa amphibia*		1	1		
18.	*Nitella opaca*				1	1
19.	*Chara ohridana*				1	1
20.	*Nitellopsis obtusa*			1		

At the researched sites of the tributaries of Lake Ohrid (River Sateska before redirection, River Sateska middle course, River Sateska inlet, River Koselska inlet and River Cherava inlet), a total of 20 macrophyte species were recorded (Tab. 1.)

Sixteen of these species belong to the group of vascular macrophytes, 3 belong to the group of Charophytes, and 1 belongs to the group of mosses. The plant abundance of the recorded species (quantitative composition of macrophytes), according to the five-point scale ranged from "very rare", "rare" to "common" (Tab. 1.).

According to the registered macrophytes, the trophic state of the researched sites was determined.

At the sites at River Sateska inlet, Koselska inlet and River Cherava inlet *Chara ohridana*, *Nitella opaca*, and *Nitellopsis obtusa* were present. Their presence indicates that the water can still be considered to be of high quality.

Elodea canadensis and *Lemna minor,* both indicators of eutrophic conditions, were registered at only one sampling site, Sateska middle course, though at low abundance (very rare). Their presence indicates an increased trophic state of the water.

The presence of eutrophic species at the tributaries of Lake Ohrid indicates that in some areas phosphorus concentrations may be elevated, leading to more eutrophic conditions.

The qualitative composition of macrophytes is an indicator of the degree of pollution of water (Sladacek, 1973; Hofrat – Ottendorfer, 1983).

The saprobity of tributaries of Lakes Ohrid and Prespa (Tab. 2.) varies from category I (oligosaprobity) at River Sateska inlet to categories I-II (oligo-metasaprobity) at River Sateska middle course, River Koselska inlet, River Cherava inlet, River Golema middle course and River Golema inlet.

Macrophytes registered at the sampling point River Sateska upper course (before redirection) are not present in both lists of the macrophyte indicators (Sladacek, 1973; Hofrat – Ottendorfer, 1983).

The data indicate that the water at the sites investigated is moderately polluted.

Table 2. The saporobity of waters in tributaries in Lake Ohrid

	Researches sites in tributaries River Sateska, River Koselska, and River Cherava	Trophic state of Lake according to physico-chemical parameters		Saprobity according to Sladecek (1973)	Saprobity according to Ottendorfer (1983)
		TP (total phosphorus)	**BOD**		
1.	River Sateska upper	high	high	-	-
2.	River Sateska middle course	high	high	O - β category (oligo-metasaprobity)	I-II category
3.	River Sateska inlet	high	high	O category (oligosaprobity)	I category
4.	River Koselska inlet	high	high	O – βcategory (oligo-metasaprobity)	I-II category
5.	River Cherava inlet	high	high	O - β category (oligo-metasaprobity)	I-II category

3. LAKE PRESPA

Lake Prespa is the second largest lake in Macedonia (Fig.1). It is situated in the south-western part at an altitude of 853 m. Lake Prespa is a transboundary lake with a surface area of 274 km^2, shared by the R. Macedonia (176.8 km^2), Albania (49 km^2) and Greece (48 km^2). Its maximum length is about 28 km, its maximum width about 17 km, and its deepest point is at 54 m. The lake is divided into two parts: Macro and Micro Prespa.

In the littoral zone of Lake Prespa, the composition of the macrophyte vegetation varies depending on water depth. Emergent and floating macrophytes grow at smaller depths and closer to the coastline while submerged macrophytes grow in deeper water.

During past decades, Lake Prespa was under a great anthropogenic pressure, which drastically affected the ecosystem, resulting in a decline of the water level and an increase of the trophic status.

3.1. Monitoring of macrophytes at Lake Prespa

Macrophytes at Lake Prespa (Fig. 3.) were monitored at the following sampling points: Ezerani, Ezerani NE, Ezerani NW, Oteshevo and Lake Prespa tributaries, mainly the River Golema inlet. The monitoring conducted at Lake Prespa includes:

- **Collection of macrophytes from the shoreline to the lower vegetation limit in the lake**
- **Preparation of a list of species found at the sampling sites (qualitative composition of macrophytes),**
- **Estimation of species abundance (quantitative composition of macrophytes) at the sampling sites,**
- **Determination of the trophic state according to the macrophytes registered, and of the macrophyte index for all investigated profiles at each depth zone.**

Figure 3. Map of Lake Prespa, including Ezerani, Ezerani NE, Ezerani NW and Oteshevo and Lake Prespa tributaries River Golema inlet

3.2. Field survey

3.2.1. Equipment used for the field survey of Lake Prespa

- **Plastic bags for the collection of samples, small hard plastic containers for fragile species (*Chara* sp. etc.) with additional waterproof labels;**
- **Field data entry sheets and field pad;**
- **Floras and relevant field guides for preliminary identification of macrophyte species;**
- **Boat suitable for local conditions;**
- **GPS (Global Positioning System)**
- **Van-Veen grab with soft rope marked for depth readings.**

3.3. Methodology

At Lake Prespa, macrophytes were collected from the shoreline to the lower vegetation limit.

For the analysis of the aquatic vegetation, the transect-based WISER method (Fig. 4.) was used. This method is recommended by the European Committee for Standardisation CEN (Comité Europeén ade Normalisation) [CEN 2002, 2003].

The WISER method consists of establishing transects perpendicular to the lake shoreline with a length covering the complete depth range of macrophyte occurrence. At each site, a central transect was selected (first of three transects per site). The other two transects were located to the left and right, respectively, of the first transect. The minimum inter-distance between two transects per site was 5 m. The starting point of the transect was situated at the beginning of the supralittoral vegetation (wetland). Transect were subdivided into 1 m depth zones (1 m depth intervals). Within each depth zone, five sampling sites were determined.

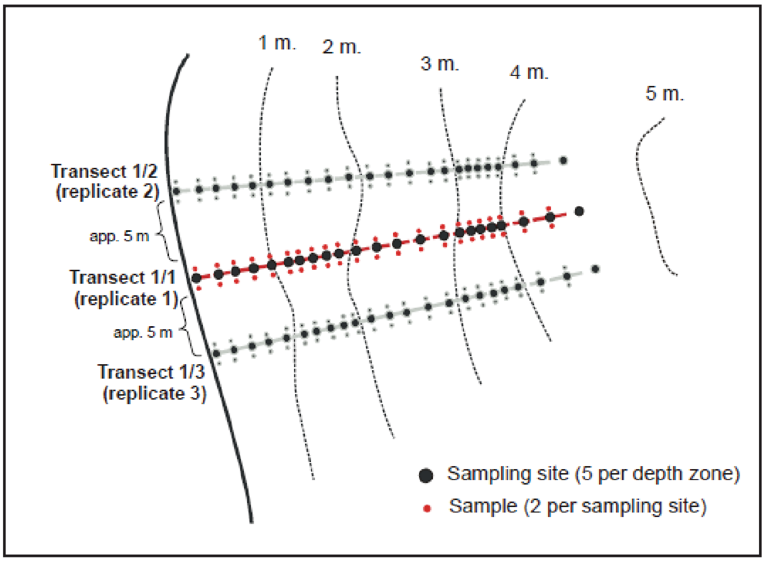

Figure 4. Sampling pattern within a sampling location (transect with three replicates)

In shallow water, entire plants were collected manually. In deep water, the plants were collected using of Van-Veen grab (Fig 5.). The plant material was washed carefully to remove sediments, epiphytes and animals.

We followed a German *Field Protocol for Macrophytes* for shores and shallow water (The assessment system PHYLIB for macrophytes and phytobenthos in running Waters and lakes for the Implementation of the EU Water Framework Directive in Germany, (2005-2008), provided by the Bavarian Environment Agency (Fig. 5.).

Specimens that could not be identified in the field were stored in plastic bags for further analyses. Bigger plants were dried on newspaper while small thin-leaved or fragile plants were preserved in alcohol.

Figure 5. Field survey at Lake Prespa

3.4. Laboratory Work

The collected material was identified to species level using different floras and keys:

- **Vascular macrophytes: Hayek, ed. (1924-1933); Jordanov, ed. (1963-1970); Josifović, ed. (1970-1977); Tutin et al., ed. (1964-1980), and**
- **Charophytes: Corillion (1957, 1975), Golerbah & Krasavina (1983), Krause (1997), Schubert & Blindow (2003) and Wood & Imahori (1964, 1965).**

Species occurrence was registered at each transect and depth zone, and the abundance of each species was estimated according to a five degree scale (Tüxen & Preising 1942 in Melzer 1999): 1 = very rare, 2 = infrequent, 3 = common, 4 = frequent, 5 = predominant (abundant)

A catalogue of nine indicator groups of macrophyte species (Melzer & Schneider, 2001), exhibiting different sensitivity towards nutrient enrichment, was used to calculate the macrophyte index, ranging from 1 to 5, with higher values indicating nutrient enrichment (Tab. 3.). Species belonging to indicator group 1 are restricted to oligotrophic conditions, whereas those belonging to indicator group 5 mainly occur in eutrophic, nutrient-rich lakes or sections of lakes. The remaining seven groups range from 1.5 to 4.5 and represent transitions between these two extremes. In addition to the MI per site, we calculated the MI separately for each depth zone at each site in order to compare the results from the shallow water with the results from the deeper water.

Table3. Macrophyte indicator groups (Melzer & Schneider, 2001)

Indicator group 1.0	**Indicator group 1.5**	**Indicator group 2.0**
Chara hispida *Chara polyacantha* *Chara strigosa* *Potamogeton coloratus* *Utricularia stygia*	*Chara aspera* *Chara intermedia* *Utricularia minor*	*Chara delicatula* *Chara tomentosa* *Potamogeton alpinus*
Indicator group 2.5	**Indicator group 3.0**	**Indicator group 3.5**
Chara contraria *Chara fragilis* *Nitella opaca* *Nitellopsis obtusa* *Potamogeton gramineus* *Potamogeton natans* *Potamogeton x zixii*	*Chara vulgaris* *Myriophyllum spicatum* *Potamogeton filiformis* *Potamogeton perfoliatus* *Utricularia australis*	*Myriophyllum verticilatum* *Potamogeton berchtoldii* *Potamogeton lucens* *Potamogeton praelongus* *Potamogeton pusillus*
Indicator group 4.0	**Indicator group 4.5**	**Indicator group 5.0**
Hippuris vulgaris *Lagarosiphon major* *Potamogeton pectinatus*	*Elodea canadensis* *Elodea nuttallii* *Potamogeton compressus* *Potamogeton crispus* *Potamogeton obtusifolius* *Ranunculus circinatus* *Ranunculus trichophyllus*	*Ceratophyllum demersum* *Lemna minor* *Potamogeton mucronatus* *Potamogeton nodosus* *Sagittaria sagittifolia* *Spirodela polyrhiza* *Zannichelia palustris*

The macrophyte index was calculated according to the formula described by Melzer (1999) but with updated indicator values and class boundaries as described in Melzer & Schneider (2001).

$$M = \frac{\sum_{i=1}^{n} I_i \cdot Q_i}{\sum_{i=1}^{n} Q_i}$$

where:

Mi = Macrophyte index

Ii = Indicator value of i-th species

Qi = Plant quantity of i-th species

n = Total number of species with an indicator value

To calculate the macrophyte index each abundance value was cubed (y D x^3) because the correlation between abundance and plant biomass is not linear (Melzer, 1999).

According to the calculation, the macrophyte index is divided into six classes, each representing different degrees of nutrient enrichment. Different colours are assigned to each of these six classes to allow a clear illustration of the results (Tab. 4).

Table 4. Relationship between index class, nutrient pollution and trophic state (Melzer & Schneider 2001)

Macrophyte index	Nutrient enrichment	Trophic state
1.00 – 2.39	very low	oligotrophic
2.40 – 2.69	low	oligo-mesotrophic
2.70 – 2.94	moderate	mesotrophic 1
2.95 – 3.30	moderate-immense	mesotrophic 2
3.30 – 3.55	immense	eutrophic 1
3.55 – 3.89	heavy	eutrophic 2
3.89 – 5.00	massive	eutrophic 3

3.5. Results and Discussion

In total, 21 macrophyte species were recorded at the research sites of the littoral zone and the tributary of Lake Prespa (Tab. 5). Six of these species are classified as helophytes and 15 a hydrophytes. No charophytes were recorded at the littoral region of Lake Prespa and the River Golema inlet site.

The dominant helophyte species at Lake Prespa is *Phragmites australis* which was present at all sites of the littoral zone and at the tributary of Lake Prespa.

In 3 sites it has been "abundant" (Ezerani, Ezerani NE, Ezerani NW), in 1 it has been "frequent" (Oteshevo) and in 1 it has been "common" (River Golema inlet). *Typha latifolia* was also present in all research sites from Lake Prespa and it was mostly characterized as "infrequent". The dominant species of hydrophytes in Lake Prespa were representatives of the Potamogetonaceae family: *Potamogeton perfoliatus, Potamogeton pectinatus* and *Potamogeton lucens*. *Potamogeton lucens* which has been recorded almost in all researched sites and they have been "abundant" in the sites Ezerani, Ezerani NE, Ezerani NW in depth of 4-5 meters, and "frequent", in site Oteshevo in depth of 1-2 meters. Also *Vallisneria spiralis* was "frequent", in site Oteshevo in depth of 1-2 meters.

In all researched sites the macroscopic algae *Cladophora* sp was present.

Table 5. Composition and abundance of macrophytes (vascular macrophytes and charophytes) from the littoral part and tributary of Lake Prespa. Numbers in bold signify indicator species according to Melzer & Schneider (2001)

	LITTORAL PART AND TRIBUTARY OF LAKE PRESPA																				
		Ezerani (littoral, 0-1m)	Ezerani (littoral, 1-2m)	Ezerani (littoral, 2-3m)	Ezerani (littoral, 3-4m)	Ezerani (littoral, 4-5m)	Ezerani (NE- littoral, 0-1m)	Ezerani (NE- littoral, 1-2m)	Ezerani (NE- littoral, 2-3m)	Ezerani (NE- littoral, 3-4m)	Ezerani (NE- littoral, 4-5m)	Ezerani (NW- littoral, 0-1m)	Ezerani (NW- littoral, 1-2 m)	Ezerani (NW- littoral, 2-3 m)	Ezerani (NW- littoral, 3-4m)	Ezerani (NW- littoral, 4-5m)	Otesevo (0-1m)	Otesevo (1-2m)	Otesevo (2-3 m)	Otesevo (3-4 m)	Lake Prespa tributary Golema (inlet)
1.	*Phragmites australis*	5	5				5	5				5	5				4				3
2.	*Typha angustifolia*	2	2				2	1				2	2				1				2
3.	*Typha latifolia*																				1
4.	*Shoenoplectus lacustris*																				1
5.	*Rumex palustris*																				1
6.	*Epilobium hirsutum*																				2
7.	*Potamogeton crispus*			1	1									1	2				3	3	
8.	*Potamogeton pectinatus*			1	1	1			1	1	2			2	1	1				3	
9.	*Potamogeton trichoides*																1			1	
10.	*Potamogeton perfoliatus*		1	2	2	2			2	2	1			2	4	2	1	2	5		
11.	*Potamogeton lucens*				1	5				1	5			5	2	5	2	4	2		
12.	*Potamogeton pusillus*																		1		

13.	*Ceratophyllum demersum*					1										1				1	
14.	*Ceratophyllum submersum*																		1		
15.	*Najas major*			1	1	2			1		2								1		
16.	*Myriophyllum spicatum*																1	3			
17.	*Vallisneria spiralis*																2	4			
18.	*Calitriche verna*																				1
19.	*Calitriche stagnalis*																				3
20.	*Ranunculus circinatus*																				3
21.	*Potamogeton natans*																				2

Submerged macrophytes are of particular importance in the aquatic ecosystems. They have well-defined ecological optima and ranges and are widely used as indicators of the trophic status of lakes and rivers (Kohler & Schneider 2003).

Contrary to water chemistry, the macrophyte index is important for the determination of trophic state because it indicates eutrophication processes in littoral ecosystems, responding to nutrient loads of both water and sediment. Macrophyte indices are considered applicable over large parts of Europe because indicator values correspond well to different indicator systems. In order to get a clearer picture of the nutrient enrichment of shallow water compared to the deeper littoral, we calculated macrophyte indices for different depth zones, notably 1-2 m, 2-3 m, 3-4 m and 4-5 m.

In the shallow water between 0 and 1 m of depth, no macrophyte index was calculated since it is defined to include only submerged or floating macrophytes (only helophytes were present - emerged macrophytes).

The values of the macrophyte index (MI) for all sampling sites from the littoral region of the Lake Prespa (Ezerani, Ezerani NE, Ezerani NW and Oteshevo) were immense - eutrophic 1 (Tab.6.).

Table 6. Trophic state of the water in researched profiles in Lake Prespa according to the MI

Sampling sites	Trophic state (physico- chemical parameters)		Macrophyte index (total)	Trophic state (MI)
	TSI (trophic stat state index)	**TP (total phosphorus)**		
Ezerani	eutrophic	good	**3.45**	immense (eutrophic1)
Ezerani NE	eutrophic	high	**3.45**	immense (eutrophic1)
Ezerani NW	eutrophic	high	**3.39**	immense (eutrophic1)
Oteshevo	eutrophic	high	**3.46**	immense (eutrophic1)

Table 7. Trophic state of Ezerani (depth points) in Lake Prespa according to the MI

Depth points	**Macrophyte Index (MI)**	**Trophic state of Ezerani according to MI**
Ezerani (0-1m)	-	-
Ezerani (1-2m)	3.00	moderate-immense (mesotrophic 2)
Ezerani (2-3m)	3.15	moderate-immense (mesotrophic 2)
Ezerani (3-4m)	3.27	moderate-immense (mesotrophic 2)
Ezerani (4-5m)	3.49	immense (eutrophic1)
Ezerani (total MI)	**3.45**	immense (eutrophic1)

Table 8. Trophic state of Ezerani NE (depth points) in Lake Prespa according to the MI

Depth points	Macrophyte Index (MI)	Trophic state of Ezerani according to MI
Ezerani (0-1m)	-	-
Ezerani (1-2m)	-	-
Ezerani (2-3m)	3.11	moderate-immense (mesotrophic 2)
Ezerani (3-4m)	3.15	moderate-immense (mesotrophic 2)
Ezerani (4-5m)	3.53	immense (eutrophic1)
Ezerani (total MI)	**3.45**	immense (eutrophic1)

Table 9. Trophic state of Ezerani NW (depth points) in Lake Prespa according to the MI

Depth points	Macrophyte index (MI)	Trophic state of Ezerani NW according to MI
Ezerani NW (0-1m)	-	-
Ezerani NW (1-2m)	-	-
Ezerani NW (2-3m)	3. 51	immense (eutrophic1)
Ezerani NW (3-4m)	3.21	moderate immense (mesotrophic2)
Ezerani NW (4-5m)	3.49	immense (eutrophic1)
Ezerani NW (total MI)	**3.39**	immense (eutrophic1)

Table 10. Trophic state of Oteshevo (depth points) in Lake Prespa according to the MI

Depth points	Macrophyte index (MI)	Trophic state of Otesevo according to MI
Oteshevo (0-1m)	3.40	immense (eutrophic1)
Oteshevo (1-2m)	3.32	immense (eutrophic1)
Oteshevo (2-3m)	3.28	moderate immense (mesotrophic2)
Oteshevo (3-4m)	4.26	massive (eutrophic3)
Oteshevo (4-5m)	-	-
Oteshevo (total MI)	**3.46**	immense (eutrophic1)

The values of the macrophyte index for all investigated sites at Lake Prespa generally indicate an immense trophic state of the water.

In shallow water, between 1 and 2 m of depth, the macrophyte index varies from moderate-immense at Ezerani to immense at Oteshevo (Tab. 7 and 10).

The macrophyte index of the 2-3 m of depth zone varies from moderate to immense: moderate at Ezerani NE, Ezerani, Otesevo and immense at Ezerani NW. At 3-4 m depth, the index varies from moderate to immense: moderate immense at Ezerani NE, Ezerani NW and Ezerani and immense at Oteshevo (Tab.7.,8.,9.,10.).

The highest values of the macrophyte index were recorded at 4-5 m depth where it varied from immense at Ezerani, Ezerani NW and Ezerani NE to massive at Oteshevo.

4. CONCLUSIONS AND RECOMENDATIONS

- **Twenty-one macrophyte species were found at the littoral zone of Lake Prespa and River Golema inlet. All these species belong to the vascular macrophytes, 6 of them are helophytes, and 15 are hydrophytes.**
- **Charophytes, which are the main indicators of clean and unpolluted water, were not found.**
- **The macrophyte index (total) for the littoral zone of Lake Prespa (Ezerani, Ezerani NE, Ezerani NW and Oteshevo) is immense (eutrophic 1).**
- **The highest values were recorded for the 4-5 m depth zone where they varied from immense (eutrophic1): Ezerani, Ezerani NW and Ezerani NE to massive (eutrophic 3) - Oteshevo.**
- **The results, however, are not representative for the whole lake because the number of transects was limited. Furthermore, some of the macrophyte species recorded are not included in the list of macrophyte indicator groups (Melzer & Schneider, 2001). It is therefore recommended to amend the existing table with groups of macrophyte indicator species that are present at the lake.**
- **Another recommendation is that more sampling sites should be studied to get a representative result for the entire lake.**
- **Measures should be taken to decrease nutrient loads in particular in areas characaterized by a high Macrophytic Index.**

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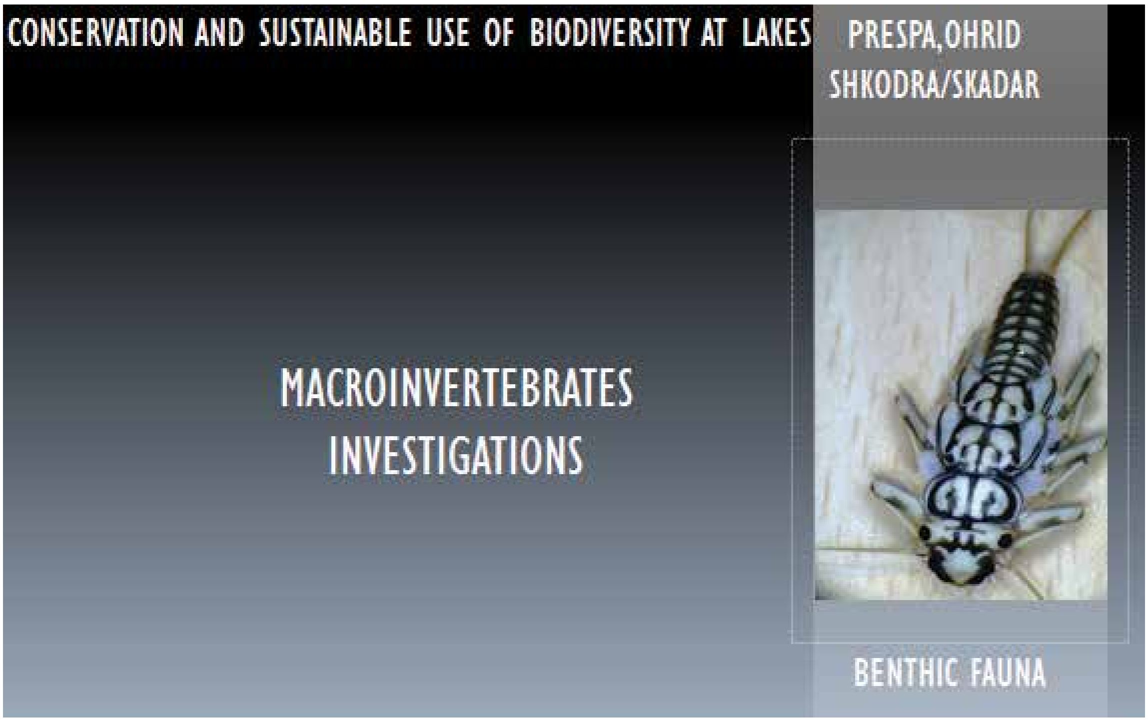

D-r Sasho Trajanovski

Ohrid, May 2014

Contents

1. INTRODUCTION

Benthic macroinvertebrates are common inhabitants of lakes and streams where they are important in moving energy through food webs. The term “benthic” means “bottom-living”, so these organisms usually inhabit bottom substrates for at least part of their life cycle; the prefix “macro” indicates that these organisms are retained by mesh sizes of ~200-500 mm (Rosenberg and Resh 1993).

Benthic macroinvertebrate species are differentially sensitive to many biotic and abiotic factors in their environment. Consequently, macroinvertebrate community structure has commonly been used as an indicator of the condition of an aquatic system (Armitage et al., 1983; Rosenberg and Resh, 1993). The use of community structure of freshwater organisms for biomonitoring can be traced back to the pioneering work of two German scientists, R. Kolkwitz and M. Marsson, in the early 1900s. Their publication on saprobity (degree of pollution) led to the development of indicator organisms. Today, indicators are widely used to summarize a wide variety of states – from biological health to economics.

There are compelling reasons for the apparent popularity of freshwater macroinvertebrates in current biomonitoring practices; they offer a number of advantages (Rosenberg et al., 1997): they are ubiquitous; they are species-rich, so the large number of species produced a range of responses; they are relatively sedentary; they are long-lived, which allows temporal changes in abundance and age structure to be followed, and they integrate conditions temporally, so, like any biotic group, they provide evidence of conditions over long periods of time.

Based on the above mentioned facts, the benthic fauna have been given a high importance in defining the ecological status of the water bodies in the European water directive. Together with 4 other biological components, the benthic fauna is listed as an obligatory biological component for the successful monitoring of the water bodies.

In the framework of the project “Conservation and Sustainable Use of the Biodiversity of Lakes Prespa, Ohrid and Shkodra/Skadar-CSBL”, macroinvertebrates were monitored at the tributaries of Lake Ohrid, one tributary of Lake Prespa and at Lake Prespa itself. The following text gives us an overview of the ongoing activities, as well as those that have been completed so far.

2. SAMPLING SITES

The sampling sites were selected based on differences in the level of anthropogenic pressures, bottom heterogeneity and the availability of data from previous studies. The latter criterion leads to the exclusion of Lake Ohrid for which plenty of data had been gathered in recent studies.

Figure1. The sampling sites (11 in total)

The role of the four tributaries (rivers Sateska, Koselska, Grasnica and Cerava), is rather important, regarding the sustaining of the water balance of Lake Ohrid. From the early 1970s until the present day, with the intensification of the industrial development in the watershed of the lake, the negative influence of the tributaries on the water quality and the habitat destruction in the littoral of the lake have been identified. However, until the latest analyses in the frame of this project, these water bodies have been given little or no attention regarding the monitoring of the living components that inhabit them. As mentioned above, the selection of sampling points was done based on differences in anthropogenic pressures in order to identify the impact of these pressures on the structure of the benthic fauna and how the fauna in turn reflects the ecological status of water and water quality. The criterion "bottom heterogeneity" was applied to distinguish anthropogenic pressures from natural conditions causing poor abundance and density of the benthic fauna.

Sampling points were selected to reflect less disturbed upstream situations in less populated areas as well as downstream situations corresponding to rural or urban populated areas. In all of the cases, one of the sampling points was before the inflow of the tributaries in order to check how the inflows influence the lower littoral of Lakes Ohrid and Prespa, respectively.

As mentioned above, samples were collected from different habitats at the sampling points so as to distinguish changes in community structure resulting from poor natural living conditions from those resulting from anthropogenic pressures. In the watershed of Lake Ohrid, the following sampling points were investigated (Fig.1.):

Lake Ohrid watershed: River Sateska: 3 points; River Koselska: 1 point; River Cerava: 1 point

Lake Prespa watershed: River Golema: 2 points

Lake Prespa littoral: Oteshevo: 3 points; NE of Ezerani: 3 points; Ezerani: 3 points; NW of Ezerani: 3 points

3. DYNAMICS AND METHODS

Following the natural laws and the life cycle of macroinvertebrates, as the most relevant, it was decided that it shall be collected twice a year: in spring and autumn. The spring and fall sampling corresponds with the highest abundance and diversity of the benthic fauna. The suggested months, May and October, correspond with the seasons when the macroinvertebrates communities reach their highest abundance and diversity. Seasonal variability of the benthic community structure and productivity is high because many species of the benthic macro invertebrates have annual (or shorter) life cycles, which culminate in an adult phase during the open water period. Therefore, it is best to sample in early spring when the larval forms are present in the sample, or in late fall, the period when most of the species have mated and the immature ones have had a chance to develop for the winter period, throughout the summer preparation. Based on the statements given above as well as the previous expertise and experience, the three partner countries have agreed to build up and adopt a joint protocol for macroinvertebrates which included the sampling methods and dynamics, lab work, data analysys and processing. Thus, it was agreed to do the sampling twice per year: Spring (late April or early May) and fall sampling (October).

The following standard limnological methods (Wetzel 1975, Wetzel&Likens 1979, Lind 1985) for collecting macroinvertebrates were used:

- Multi habitats transect method at Lake Prespa, using a van Veen grab
- Kick and swipe in the tributaries, using a D-shaped net.

A B

Figure 2. Sampling equipment: van Veen grab and D-shaped net

A multi-habitat transect-line sampling method was used for collecting macroinvertebrates at Lake Prespa, using a Van Veen grab (Fig.2A) according to standard methods for macroinvertebrates sampling. At least 3 replicate samples were taken in each sampling point. The samples were taken from three depth points in each transect. The deepest sampling point was the lowest depth limit of macrophyte vegetation. The samples were collected using the direction from the shore to the deeper zone (vertically).

The kick and sweep method (ISO: EN 27828:1994 AQEM/STAR-lakes: Cheshmedjiev et al., 2011) was used during the sampling of rivers on sandy, gravelly and mixed bottoms covered with macrophyte vegetation. The kick net is D-shaped, with a metal frame holding a mesh bag of 400-mm size (Fig. 2B). The standard kicking time interval was 5 minutes. In order to get the qualitative composition of the sampled area, a metal rectangular square (1m^2 square) is set on the bottom where the sampling is carried out. This sampling type has been used on all sampling transects in depth points of 0.5 m. The samples have been sieved, preserved with 70% ethanol and transported to the Laboratory for further examination.

The determination was done using the following keys: Lukin 1976, Sapkarev 1964, Radoman 1983; 1985, Kerovec 1986, Polinski (1929), Snegarova (1954), Radoman (1959), Hubendick (1960, 1970), Hadzisce (1974), Sapkarev (1964), Krstanovski (1994) etc.

The main goal in the study was to monitor macroinvertebrates as a quality element according to the European Water Framework Directive (WFD 2000), considering the following parameters and indices:

- Benthic community structure: species composition (diversity) and density (abundance).
- Ecological status: IBI, ASPT, Shannon & Wiener Diversity index, Margalef, Simpson, Pielou etc., (Mandaville, 2002; Merritt, 1996; Plafkin, 1989).

Having in mind that water monitoring in accordance to the EWFD doesn't have long tradition in Macedonia as well as both water bodies Lake Prespa and Lake Ohrid are very unique lakes, choosing a relevant index for assessing the ecological status was very sensitive issue and was given special attention. During the process of harmonization of the methods and data analysis, the teams have agreed to use ASPT index (Average Score Per Taxon) for assessing the ecological status of the sampling sites in the Lakes, i.e. IBI (originally Biotic Index by William M. Beck) for assessment of the ecological status of the sampling sites in the rivers (Macedonian partner only).

The ASPT index has been widely used for assessment of the ecological status of the European water bodies especially in the frame of WISER project which support the implementation of the Water Framework Directive (WFD) by developing tools for the integrated assessment of the ecological status of European surface waters (wiser.eu). It is based on the biological monitoring working party (BMWP)- a procedure for measuring water quality using the macroinvertebrates (up to level of families) sensitivity to different pollutants. It has been initially developed by Armitage (Armitage et al, 1983). The scoring values starts from 1 which indicates lowest quality of the water (Oligochaeta) to 10 (stoneflies and mayflies) indicating very clean water bodies. The ASPT equals the average of the tolerance scores of all macroinvertebrate families found, and ranges from 0 to 10.

BI-the Biotic Index (or modified Irish Biotic Index, Cheshmedziev, 2011) was originally developed for streams. It has successfully been applied in many European countries, including Bulgaria, during the process of assessing the quality of the water bodies under the European WFD. The geographical position (similarity) of two neighboring countries (both Macedonia and Bulgaria are Balkan countries) and the positive experience with the use of BI in Bulgaria, has key role in choosing of this index in assessment of the water quality in the rivers in the watershed of Prespa and Ohrid lakes. The biotic index works by assigning different levels of tolerance to pollution to the different types of organisms. The types of macroinvertebrates found during sampling are divided into 4 groups: The final scoring scale starts from 1-5 corresponding with 5 basic level of pollution (high, good, moderate, poor and bad).

A try to use the traditional metrics (Shannon & Wiener Diversity index, Margalef, Simpson, Pielou etc) in assessing the ecological status of the water bodies was done by extrapolating of the obtained values in the system of values between referent and bad ecological conditions, developed in Bulgaria (Project 563, 2004). Comparing the results from IBI with the results from the traditional metrics, it is obvious that this system is not always "following" the values of IBI, or the real in situ status of the water bodies, a fact that gives an advantage to IBI and ASPT over traditional metrics in assessing the ecological status.

For all calculated indexes, for more comprehensive interpretation of the ecological status, there have been used the appropriate colors in accordance to the colorful scale of European WFD: red= bad status; orange=poor status, yellow=moderate status, green=good and blue=high ecological status (See Tables 7, 15 and the Tables in Appendix).

4. RESULTS AND DISCUSSION

The following results were obtained during the examination of the macroinvertebrate fauna in spring and fall 2014.

During the spring sampling campaign (25, 30 May 2013), 15 samples were collected from the tributaries of Lake Ohrid (River Sateska, Koselska and Cerava), 6 from River Golema and 36 from the littoral localities along Lake Prespa. Therefore, a total of 56 samples were collected during the spring campaign, whereby the community structure based on the taxonomical determination of the species has been determined.

4.1. Lake Ohrid watershed

4.1.1. River Sateska

River Sateska is one of the most important tributaries of Lake Ohrid. Its watershed covers about 39% of the watershed of the lake. It is not a natural tributary of Lake Ohrid, but in order to reduce the sediment transport of Crni Drim and to facilitate the regulation of its discharge, in 1962, the river was diverted into the Lake (Jordanoski et al., 2007).

The River Sateska flows through both agricultural and urban areas and carries a very high load of both phosphorus and sediment, which is deposited in the shallow waters of Lake Ohrid at the mouth of the river. The load of phosphorus coming from the Sateska River may be about the same as that coming from the sewerage of Pogradec (Watzin et al., 2002).

Figure 3. River Sateska Upper Course

The macrozoobenthic communities have been researched in River Sateska by taking samples from three sites along the river: Upper course, in the upper flow; middle course in the middle flow and inflow close to the inflow into the Lake.

4.1.2. River Sateska Upper Course

4.1.2.1 Spring Sampling Campaign

Table 1. Diversity and density of the macroinvertebrates from the Upper Course of River Sateska in spring 2014.

Locality: River Sateska Upper Course Coordinates: 41°14'26.85"N; 20°45'50.63"E			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Glossiphonia complanata*	1	25
Gastropoda	*Ancylus fluviatilis*	1	25
Amphipoda	*Gammarus balcanicus*	36	900
Insecta	*Baetis vernus (Ephemeroptera)*	32	800
	Epeorus pleuralis (Ephemeroptera)	4	100
	Ephemera danica (Ephemeroptera)	17	425
	Ephemerella sp. (Ephemeroptera)	6	150
	Ecdyonurus venosus (Ephemeroptera)	4	100
	Rhithrogena sp. (Ephemeroptera)	23	575
	Leuctra ferruginea (Plecoptera)	1	25
	Isoperla sp. (Plecoptera)	1	25
	Perla marginata (Plecoptera)	14	350
	Limnephilus sp. (Trichoptera)	1	25
	Sericostoma sp. (Trichoptera)	6	150
	Silo sp. (Trichoptera)	3	75
	Hydropsyche sp.(Trichoptera)	1	25
	Leptocerus sp. (Trichoptera)	49	1225
	Atheryx sp. (Diptera)	3	75
	Tabanus sp. (Diptera)	1	25
	Tanypus sp. (Diptera)	1	25
	Bezzia sp. (Diptera)	1	25
4	**21**	**206**	**5150**

The Upper flow of River Sateska is without any anthropogenic impact (Fig.3.) and it could be observed as a reference site concerning the macrozoobenthic species composition.

Table 1 indicates a diverse and rich structure of the macroinvertebrates. In total, 21 taxa have been identified belonging to 4 systematic groups: Hirudinea, Gastropoda, Amphipoda and Insecta. As expected, the richest is the class of Insecta: 18 species or 90 % of the total species number belong to the class of Insecta. Most of them are characteristic for clean waters, especially the representatives from the class of Plecoptera *(Perla marginata, Isoperla sp.*). In the samples, no endemic species for Lake Ohrid and its watershed have been registered. All species are cosmopolitan. The highest densities are noticed in the populations of *Leptocerus sp.,* (Fig.5.) *Gammarus balcanicus* and *Baetis vernis.*

The values of IBI are the highest compared to all others researched localities reaching the value of 5, thus indicating a high ecological status.

4.1.2.2 Fall Sampling Campaign

Table 2 shows the structure of the macroinvertebrates in River Sateska Upper Course during the fall sampling campaign.

Table 2. Diversity and density of the macroinvertebrates from the Upper Course of River Sateska in Fall 2014.

Locality: River Sateska Upper Course Coordinates: 41°14’26.85”N; 20°45’50.63”E			
Class	Species (Total Number)	Ind	Total ind/m^2
Oligochaeta	*Criodrilus lacuum*	1	25
	Potamothrix hammoniensis	1	25
Gastropoda	*Ancylus fluviatilis*	2	50
Amphipoda	*Gammarus balcanicus*	47	1175
Insecta	*Ephemera danica (Ephemeroptera)*	33	825
	Baetis vernus (Ephemeroptera)	39	975
	Epeorus pleuralis (Ephemeroptera)	8	200
	Ecdyonurus venosus (Ephemeroptera)	5	125
	Perla marginata (Plecoptera)	21	525
	Isoperla sp. (Plecoptera)	3	75
	Sericostoma sp. (Trichoptera)	16	400
	Leptocerus sp. (Trichoptera)	49	1225
	Tipula sp.(Diptera)	4	100
	Chironomus plumosus (Diptera)	13	325
4	**14**	**242**	**6050**

14 species were registered during the fall sampling in the Upper Course of River Sateska. Same as during spring, they belong to 4 taxonomic groups, but unlike during spring, instead of Hirudinea, now the predominant class was Oligochaeta In fact, 72 % of the species belong to the class of Insecta. The presence of Plecoptera, which are highly sensitive to polluted waters, indicate clean water, as it was the case during the spring period. Again, Leptocerus sp. was the most abundant species

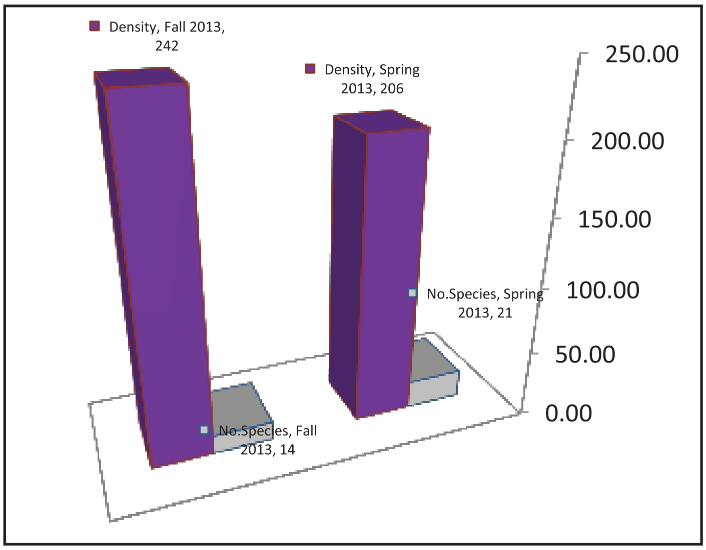

Figure 4. Comparison: density vs diversity in the Upper Course of R. Sateska (Spring vs Fall)

Figure 4 shows the comparisons between the density and diversity (in total) and the IBI values, in two different seasons. Clearly the density is higher in fall, while the diversity is higher in spring. This could be explained by the possibility that more species survived the winter conditions in a larval phase to be present in the samples, while the higher density in fall is a result of the better living conditions and the food availability in the period between two sampling campaigns. The values of IBI stay unchanged during the whole period, and they indicate a high ecological status.

As it was mentioned above, in both seasons, the highest contribution to the total diversity of the macroinvertebrates has been "given" by the representatives from Insecta. The most abundant species in both seasons was *Leptocerus sp.* (Fig.5.)

Figure 5. The most abundant species in both seasons in Upper Course of R.Sateska: Leptocerus sp.

4.1.3. River Sateska Middle Course

The samples have been taken before the river's redirection in order to assess the influence of both the diffuse and the point source pollution.

4.1.3.1 Spring Sampling Campaign

Table 3. Qualitative and quantitative composition of the macroinvertebrates in the Middle Course of River Sateska in spring 2014.

Locality: River Sateska Middle Course Coordinates: 41°12'4.63"N; 20°44'33.26"E			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Erpobdella octoculata*	4	100
Amphipoda	*Gammarus roeseli*	64	1600
Gastropoda	*Theodoxus fluviatilis*	3	75
Insecta	*Baetis vernus(Ephemeroptera)*	4	100
	Limnephilus sp. (Trichoptera)	4	100
	Tanypus sp. (Diptera)	24	600
	Atheryx sp. (Diptera)	3	75
	Tabanus sp. (Diptera)	1	25
	Gomphus vulgatissimus (Odonata)	2	50
	Limnius sp. (Coleoptera)	4	100
	Aphelocheirus aestivalis (Heteroptera)	2	50
4	**11**	**115**	**2875**

After the redirection, along the river course to its inflow into the Lake, the River passes through the populated as well as the agricultural area and it collects both the waste water from the households and the sediments washed out from the agricultural areas, together with the chemicals used during the agricultural activities.

Table 3 shows the diversity and the density of the macroinvertebrates in the Middle Course of River Sateska. It is obvious that both the density and diversity are lower compared to h the Upper Course and the Inflow of River Sateska. Here, the species from the class of Plecoptera are absent, while the majority of the species identified indicate a certain level of pollution. The total number of species is 11, belonging to 4 systematic classes: Hirudinea, Gastropoda, Amphipoda and Insecta. 8 species (73%) belong to the class of Insecta. *Gammarus roeseli* (Fig.7.) have the highest density, followed by *Tanypus sp.,* a representative from Chironomidae and an indicator of slightly polluted waters.

The values of IBI are lower in the Middle Course of River Sateska compared to the values obtained in the Upper Course, and indicate a moderate ecological status.

4.1.3.2 Fall Sampling Campaign

Table 4. Diversity and density of the macroinvertebrates from the Middle Course of River Sateska in fall 2013.

Locality: River Sateska Middle Course Coordinates: 41°12'4.63"N; 20°44'33.26"E			
Class	Species (Total Number)	Ind	Total ind/m²
Oligochaeta	*Potamothrix hammoniensis*	2	50
Hirudinea	*Erpobdella octoculata*	4	100
Amphipoda	*Gammarus roeseli*	170	4250
	Gammarus balcanicus	27	675
Gastropoda	*Ancylus fluviatilis*	1	25
Insecta	*Ephemera danica (Ephemeroptera)*	94	2350
	Baetis scambus (Ephemeroptera)	7	175
	Baetis rhodani (Ephemeroptera)	3	75
	Baetis vernis (Ephemeroptera)	30	750
	Ecdyonurus venosus (Ephemeroptera)	14	310
	Leuctra ferruginea (Plecoptera)	2	50
	Isoperla sp.(Plecoptera)	3	75
	Goeridae (Trichoptera)	2	50
	Rhyacophila sp. (Trichoptera)	1	25
	Limnephilus sp. (Trichoptera)	2	50
	Leptocerus sp. (Trichoptera)	111	2775
	Chironomus plumosus (Diptera)	3	75
	Tabanius sp. (Diptera)	4	100
	Ormosia sp.(Diptera)	1	25
	Dytiscus sp.(Coleoptera)	1	25
	Limnius sp.(Coleoptera)	1	25
5	**21**	**483**	**12075**

The table above shows a high diversity of the macroinvertebrates in the Middle Course of River Sateska. From the total of 21 species, belonging to 5 systematic groups, 76 % belong to the class of Insecta. The population of the species *Gammarus roeseli* experiences the highest density, followed by *Leptocerus sp.* and *Ephemera danica*. The species composition which includes only one representative from the class of Oligochaeta and one from Chironomidae (which are an indicator of polluted waters) indicate clean waters. The value of IBI is between 4-5 and indicates a high ecological status.

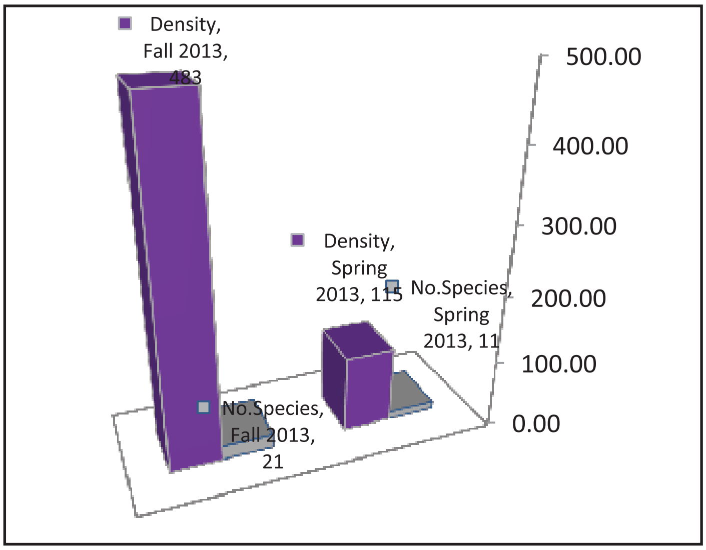

Figure 6. Comparison: density vs diversity in the Middle Course of R. Sateska (Spring vs Fall)

The comparisons between the density, diversity and IBI values in both seasons are given in Figure 6. The diversity of the macroinvertebrates in the fall is two times higher, while the density is three times higher than in spring. Also, the value of the IBI in fall is higher than in spring. The higher density and diversity are in accordance with the natural seasonal fluctuation of the macroinvertebrates and are connected to the food availability and the optimal environmental conditions that enable the development of the macrozoobenthic communities. The higher IBI values in fall could be related to the decreased level flow in the river, and the reduced agricultural activities as well as the reduced sediment loading due to the mostly dry summer period. In both seasons the highest densities have been registered in the population of *Gammarus roeseli* (Fig.7.).

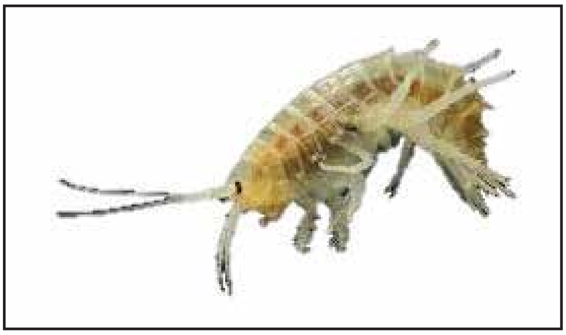

Figure 7. The most abundant species in both seasons in the Middle Course of R.Sateska: Gammarus roeseli.

4.1.4. River Sateska Inflow

4.1.4.1 Spring Sampling Campaign

Concerning the diversity in the Inflow of River Sateska 12 species have been identified, belonging to 3 classes from the macroinvertebrates. The class of Insecta comprises the highest number of the recorded species, reaching 75%. Quantitatively, the highest density (Tab.5.) has been recorded in the population of *Gammarus roeseli* (83 ind/m^2), followed by *Ephemera danica,* then *Leptocerus sp.*, etc.

As it is shown in Table 5, most of the species are typical for moderately polluted waters, including the presence of the species belonging to the class of Hirudinea. The IBI (Irish Biotic Index) reaches a value of 3 which indicates the same ecological status assessed in the Middle Course, but weaker than the Upper Course where it reached its highest value i.e. a high ecological status.

Table 5. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Sateska in spring 2014.

Locality: River Sateska Inflow Coordinates: 41°10'4.38"N; 20°43'36.59"E			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Glossiphonia complanata*	1	25
	Erpobdella octoculata	3	75
Amphipoda	*Gammarus roeseli*	83	2075
Insecta	*Ephemera danica (Ephemeroptera)*	38	950
	Ecdyonurus venosus (Ephemeroptera)	3	75
	Leuctra ferruginea (Plecoptera)	1	25
	Limnephilus sp. (Trichoptera)	3	75
	Sericostoma sp. (Trichoptera)	2	50
	Leptocerus sp.(Trichoptera)	12	300
	Limnius sp. (Coleoptera)	1	25
	Gomphus vulgatissimus (Odonata)	1	25
	Aphelocheirus aestivalis (Heteroptera)	1	25
3	**12**	**149**	**3725**

4.1.4.2 Fall Sampling Campaign

The biodiversity of the macroinvertebrates in the fall period in the Inflow of River Sateska is relatively poor. 9 species have been registered belonging to 2 systematic groups: Amphipoda and Insecta. 8 species belong to the class of Insecta while only one belongs to Amphipoda. The highest density has been recorded in the population of *Gammarus roeseli,* same as during the research in the spring period. There are no highly sensitive species from the class of Insecta, which is indicative of a water with lower quality.

Table 6. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Sateska in Fall 2014.

Locality: River Sateska Inflow Coordinates: 41°10'4.38"N; 20°43'36.59"E			
Class	Species (Total Number)	Ind	Total ind/m^2
Amphipoda	*Gammarus roeseli*	375	9375
Insecta	*Ephemera danica (Ephemeroptera)*	29	725
	Leptophlebia sp. (Ephemeroptera)	6	150
	Sericostoma sp. (Trichoptera)	3	75
	Goeridae (Trichoptera)	1	25
	Leptocerus sp. (Trichoptera)	1	25
	Chironomus plumosus (Diptera)	4	100
	Gomphus vulgatissimus (Odonata)	2	50
	Corixa sp. (Heteroptera)	1	25
2	**9**	**422**	**10550**

When comparing the density, diversity, and IBI values in spring and fall, it is obvious (Fig.8.) that the density has increased in the fall, while the diversity has decreased from 12 in the spring to 9 species in the fall. The value of the IBI has also dropped, from 3 which indicates poor ecological status to 2, which indicates bad ecological status. Since on the way to its inflow into the Lake the River passes through the populated areas, and due to the lower water flow, it is possible that the waste water and the sewage from the households could cause deterioration of the water quality in the inflow of River Sateska. The other difference in the species composition is due to the shift of the class of Hirudinea which was present in the spring with the class of Amphipoda.

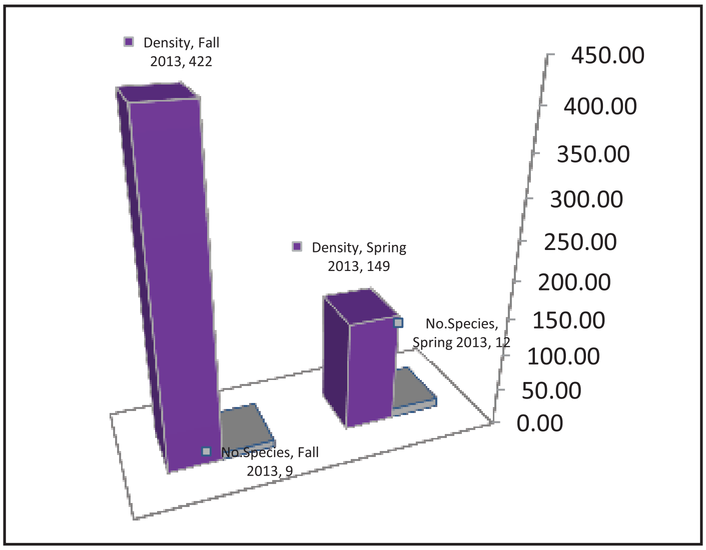

Figure 8. Comparison: density vs diversity in the Inflow of R. Sateska (Spring vs Fall)

A

B

Figure 9. IBI (Irish Biotic Index) values along the River Sateska: A-Spring; B-Fall.

Figure 9 A, shows how the Irish Biotic Index (IBI) changes along the River Sateska flow in the spring period. Going from the upper flow to the inflow, the IBI is getting worse, which corresponds with the level of anthropogenic impact along the river course.

Table 7. Values of the "traditional" metrics and IBI values along River Sateska (both seasons)

River Sateska	**SU (Spring)**	**SU (Fall)**	**SM (Spring)**	**SM (Fall)**	**SI (Spring)**	**SI (Fall)**
Number of individuals - N	206	242	115	483	149	422
Number of taxa - S	21	14.0	11	21.0	12	9.0
Diversity Shannon & Weaner - H	3,3	3.1	2,15	2.7	1,93	0.7
Species richness po Margalef - d	8,64	5.5	4,85	7.5	5,06	3.0
Species evenness Pielou - e	0,75	0.8	0,62	0.6	0,54	0.2
Species dominance index Simpson - c	0,14	0.1	0,36	0.2	0,38	0.8
IBI-Irish Biotic Index	5	5	3	4	3	2

In the Upper Course, the conditions observed indicate almost nonimpacted sites, while going down the river, the conditions change both in hydromorphology of the river bed and the intensity of the anthropogenic impact. All those have impacted the structure of the macrozoobenthic communities resulting in a presence of species typical for such living conditions. Figure 9 B, shows the values of the IBI along the River Sateska in the fall period. The upper course is still with a high ecological status, which becomes moderate in the middle course and bad in its inflow into the Lake.

The assessment of the ecological status is more or less the same, using the traditional metrics: Shannon & Weaner diversity index, Margalef species richness, species evenness- Pielou and Simpson's species dominance index (Tab.7).

4.1.5. River Koselska

4.1.5.1 Spring Sampling Campaign

The River Koselska rises in the southern slopes of Mountain Ilinska. It is a 26 km long river, and an important tributary of Lake Ohrid. Until the village of Kosel, it is a mountainous stream, well saturated with oxygen, belonging to the Ist category. From Kosel to its inflow it passes through populated areas and it receives the waste of the domestic waters from the households and sediments from the agricultural areas (Fig.10.) which contribute to deterioration of the River's water quality.

The macroinvertebrates in River Koselska was researched only in one sampling site, in the Inflow of River Koselska into the Lake. In general, both the diversity and density of the macroinvertebrates are poor. From the total of 11 species, 6 species (55%) belong to the class Insecta. Besides Insecta, two more classes have been registered: Oligochaeta and Hirudinea. The species composition is very indicative (Oligoacheata species, and Chironomidae) of aquatic ecosystem under strong anthropogenic influence. The highest density has been recorded in the population of Chironomid larvae which are typical representatives of polluted waters.

Figure 10. River Sateska-Inflow into Lake Ohrid

Table 8. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Koselska in spring 2014.

Locality: River Koselska Inflow Coordinates: N 40°56'916"; E 020°46' 463"			
Class	Species (Total Number)	Ind	Total ind/m²
Oligochaeta	*Limnodrilus hoffmeisteri*	1	25
	Stylodrilus sp.	7	175
Hirudinea	*Erpobdella octoculata*	2	50
Insecta	*Ephemera danica (Ephemeroptera)*	1	25
	Baetis vernus (Ephemeroptera)	8	200
	Caenis macrura (Ephemeroptera)	1	25
	Chironomus plumosus (Diptera)	10	250
	Hermetia sp. (Diptera)	1	25
	Ormosia sp. (Diptera)	1	25
3	**9**	**32**	**800**

The value of the Irish Biotic Index is 2, pointing to bad ecological status.

4.1.5.2 Fall Sampling Campaign

Table 9. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Koselska in fall 2014.

Locality: River Koselska Inflow Coordinates: N 40°56'916"; E 020°46' 463"			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Erpobdella octoculata*	3	75
Oligochaeta	*Potamothrix hammoniensis*	8	200
Gastropoda	*Lymnaea stagnalis*	3	75
Isopoda	*Asellus aquaticus*	1	25
Insecta	*Baetis rhodani (Ephemeroptera)*	3	75
	Sericostom sp (Trichoptera)	1	25
	Chironomus plumosus (Diptera)	18	450
5	**7**	**37**	**925**

7 species have been identified in the Inflow of River Koselska in the fall period. They belong to 5 systematic groups: Oligochaeta, Hirudinea, Gastropoda, Isopoda and Insecta. The highest biodiversity (3 species) belongs to the class of Insecta. Here, the highest density is noticed in the population of *Chironomus plumosus*, which is an indicator of polluted waters. The other species indicate polluted waters as well. The value of the Irish Biotic Index is 2, which indicates bad ecological status.

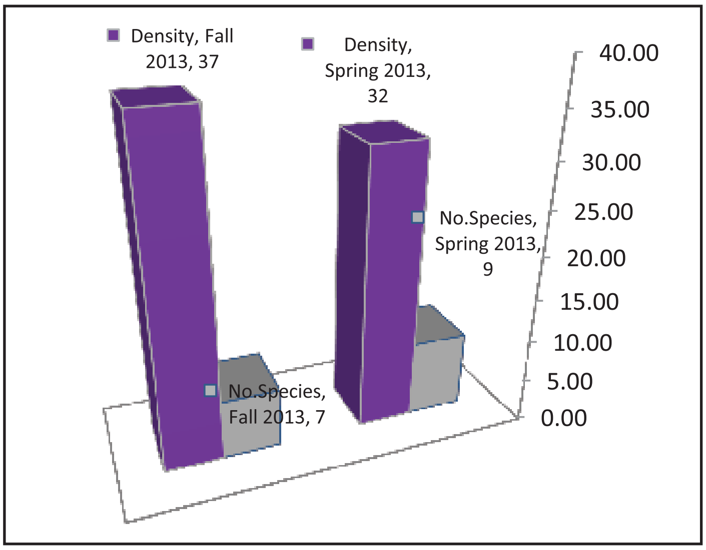

Figure 11. Comparison: density vs diversity in the Inflow of R. Koselska (Spring vs Fall)

The Figure 11 shows the comparison between the density, diversity and IBI index values. Concerning the density and IBI values, they are almost the same during both seasons. The diversity is a little higher, on the level of taxa, during the spring period while, on a higher systematic level, the diversity is higher in the fall season. In both seasons, *Chironomus plumosus* is the most abundant. This leads us to the conclusion that the structure of the benthic fauna has not been affected by the season.

4.1.6. River Cerava

The River Cerava is a tributary of Lake Ohrid in its southern part, known as one of the biggest polluters of the Lake. It rises in Albania and on the way to its inflow it passes through the populated areas where it receives the domestic waste and sewage waters, as well as solid waste. Due to the erosive character of the ground, the river water is mostly colored yellow-brown throughout the whole year. Due to these reasons, the River's water belongs to the IIIrd and IVth category (Sarafiloska et al, 2007)

Figure 12. R. Cerava

4.1.6.1 Spring Sampling Campaign

The macroinvertebrates in River Cerava was examined only in one sampling site-in the Inflow into the Lake.

Table 10 shows the diversity and the density of the macroinvertebrates in the spring time in the inflow of River Cerava. Both the diversity and density of the macroinvertebrates are poor. 7 taxa have been registered belonging to the classes of Hirudinea and Insecta. The highest density is among the family of Chironomidae, whose representatives indicate polluted waters. The endemic species have not been registered in the Inflow of River Cerava.

Table 10. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Cerava in spring 2014.

Locality: River Cerava Inflow Coordinates: N 41°55'25"; E 020°45' 21"			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Haemopis sanguisuga*	2	50
Insecta	*Tipula sp. (Diptera)*	2	50
	Chironomus sp. (Diptera)	64	1600
	Platambus sp. (Coleoptera)	2	50
	Platambus maculatus (Coleoptera)	2	50
	Aeshna grandis (Odonata)	1	25
	Coenagrion sp.(Odonata)	1	25
2	7	**74**	**1850**

The value of the Irish Biotic Index are low as well-2. This value indicates bad ecological status.

4.1.6.2 Fall Sampling Campaign

Table 11. Qualitative and quantitative composition of the macroinvertebrates in the Inflow of River Cerava in fall 2014.

Locality: River Cerava Inflow Coordinates: N 41°55'25"; E 020°45' 21"			
Class	Species (Total Number)	Ind	Total ind/m²
Amphipoda	*Gammarus roeseli*	7	175
Insecta	*Gammarus ochridensis*	8	200
	Ephemera danica (Ephemeroptera)	7	175
	Baetis vernis (Ephemeroptera)	4	100
	Sericostoma sp. (Trichoptera)	5	125
2	**5**	**31**	**775**

The biodiversity of the macroinvertebrates in the inflow of River Cerava in the fall season is poor, as it was during the spring sampling campaign. 5 species identified belong to 2 systematic groups: Amphipoda and Insecta. The only species from the Amphipoda, *Gammarus ochridensise*, is endemic to Lake Ohrid. The densities of all species are low and indicative of water environments with poor life conditions. The value of IBI is 2, and it clearly indicates a bad ecological status. The comparison between the density, diversity and IBI values is shown in Figure 13. The values of the IBI are common for both seasons. In both seasons its value is 2, which is indicative of bad ecological conditions. Concerning the biodiversity and density, both are higher in spring, during the higher water flow. The species composition where Insecta species predominate could be the explanation for both the higher density and the diversity in spring. Most of the Insecta species survive the winter conditions in the form of larvae and the spring period is a period for their maturation, according to their life cycle.

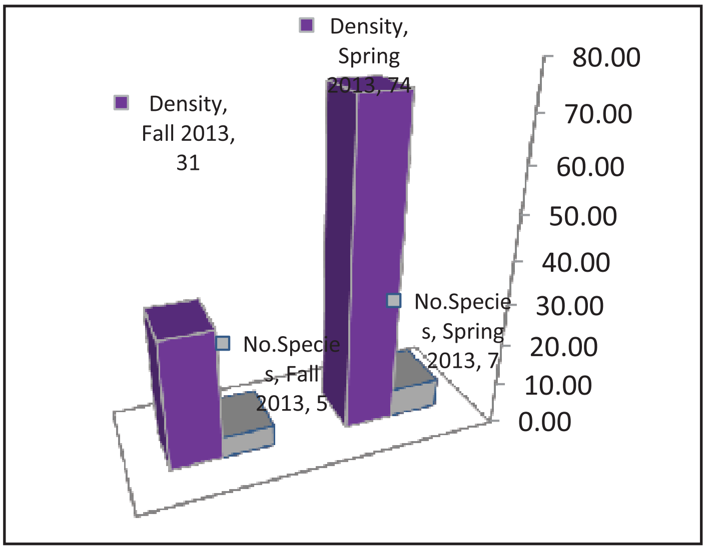

Figure 13. Comparison: density vs diversity in the Inflow of R. Cerava (Spring vs Fall)

4.2. Lake Prespa watershed

4.2.1. River Golema

The River Golema is the largest and the most important river in the Lake Prespa watershed with a total catchment area of 162 km². It covers a wide range of different areas from mountains to lowland wetlands in the delta area. The latest analyses show a deteriorated water quality due to the pressures coming from the settlements and form human activities. The main sources of pollution of the river are wastewaters produced by households, industries and agriculture (Prespa Watershed Management Plan, 2013; Fishery plan of Prespa Lake, 2008).

4.2.2. Spring Sampling Campaign

Two sampling sites have been selected for the collection of samples from the macroinvertebrates: River Golema Middle Course and River Golema Inflow into the Lake Prespa.

Table 12. Qualitative and quantitative composition of the macroinvertebrates in River Golema Middle Course in spring 2014.

Locality: River Golema Middle Coordinates: 41° 4’44.29”N; 21° 1’25.36”E			
Class	Species (Total Number)	Ind	Total ind/m²
Insecta	*Tanypus sp. (Diptera)*	35	875
	Chironomus plumosus (Diptera)	76	1900
1	**2**	**111**	**2775**

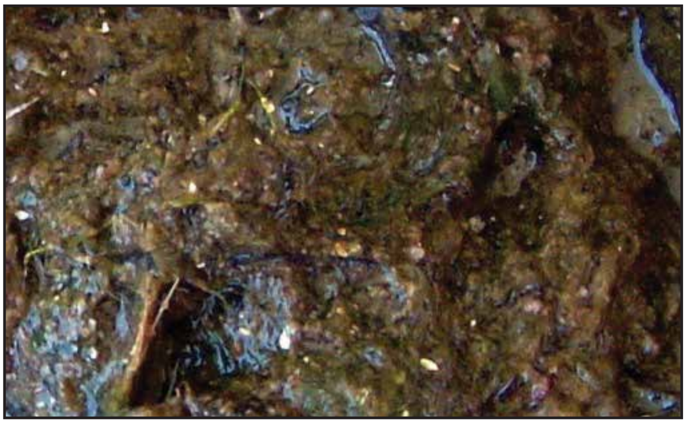

Figure 14. Bottom facies at Golema River Middle Course.

It is obvious that despite the poor diversity, both species reach very high densities, which is typical for these species in absence of competition and due to their high tolerance to pollution. Only two species have been identified, both indicators of heavy pollution. The population of *Chironomus plumosus* predominated quantitatively.

A lot of organic waste was present in the river (Fig.14.), which made it impossible to reach the bottom of the river.

The specific smell of decomposing organic waste alongside the presence of the solid waste completed the “picture” of this sampling site. The value of the Irish Biotic Index was 1, the lowest among all river sampling sites, indicating bad ecological status.

4.2.2.1 Fall Sampling Campaign

Table 13. Qualitative and quantitative composition of the macroinvertebrates in River Golema Middle Course in fall 2014.

Locality: River Golema Middle Coordinates: 41° 03’17.421”N; 21° 02’2.299”E			
Class	Species (Total Number)	Ind	Total ind/m²
Oligochaeta	*Limnodrilus hoffmeisteri*	17	425
Insecta	*Chironomus plumosus (Diptera)*	15	375
2	**2**	**32**	**800**

The table shows a very poor qualitative composition of the macroinvertebrates identified in the Middle Course of River Golema during the fall period. As it was the case during the spring, again, only two species were present in the samples. Both were quantitatively well distributed, and both were indicators of a heavy polluted water environment. The value of IBI is 1, and it clearly indicates a bad ecological status.

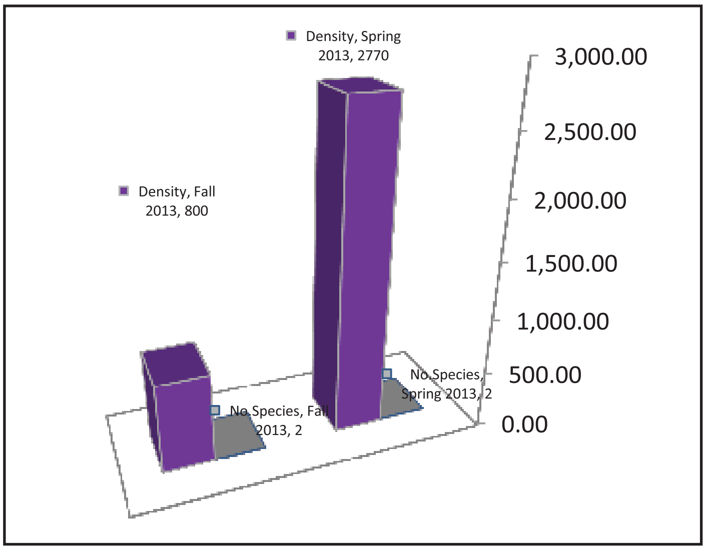

Figure 15. Comparison: density vs diversity in the Inflow of R. Golema Middle Course (Spring vs Fall)

The comparison between the density, diversity and IBI values are shown in Figure 15. The density is almost 4 times higher in spring, while the diversity stays the same during both seasons. The values of IBI are lowest among all sampling sites, and both indicate a bad ecological status. As it was mentioned above, a lot of organic waste (mainly apple pulp) as well as the solid waste create an almost anoxic life condition, where only the highest tolerant species could survive.

4.2.3. River Golema Inflow

4.2.3.1 Spring Sampling Campaign

The structure of the macroinvertebrates recorded in the Inflow of River Golema was slightly "richer" compared to the sampling site River Golema Middle Course. 5 systematic groups completed the overall diversity of this sampling site: Oligochaeta, Hirudinea, Gastropoda, Bivalvia and Insecta, each of them represented by one species except the class of Oligochaeta where two species have been registered. The presence of of *Planorbis prespensis* and *Pisidium casertanum* indicate the influence of the closeness of the Lake, since both species are typical inhabitants of Lake Prespa. A far higher density has been registered in the population of *Chironomus plumosus.*

Table 13. Qualitative and quantitative composition of the macroinvertebrates in River Golema Inflow in spring 2014.

Locality: River Golema Inflow Coordinates: 40°59'37.50"N; 21° 1'47.46"E			
Class	Species (Total Number)	Ind	Total ind/m^2
Hirudinea	*Erpobdella octoculata*	1	25
Insecta	*Chironomus plumosus*	57	1425
Bivalvia	*Pisidium casertanum*	1	25
Oligochaeta	*Potamotrix hammoniensis*	14	350
	Limnodrilus hoffmeisteri	14	350
Gastropoda	*Planorbis prespensis*	2	50
5	**6**	**89**	**2225**

The value of the Irish Biotic Index is the same as the one registered at the sampling site River Golema Middle Coruse-1 and indicates bad ecological status.

4.2.3.2 Fall Sampling Campaign

Table 14. Qualitative and quantitative composition of the macroinvertebrates in River Golema Inflow in fall 2013.

Locality: River Golema Inflow Coordinates: 40°59'37.50"N; 21° 1'47.46"E			
Class	Species (Total Number)	Ind	Total ind/m²
Oligochaeta	*Potamotrix hammoniensis*	50	1250
Hirudinea	*Erpobdella octoculata*	3	75
Gastropoda	*Planorbis prespensis*	2	50
Bivalvia	*Dreissena presbensis*	8	200
Amphipoda	*Gammarus triacanthus prespensis*	9	225
Insecta	*Chironimus plumosus*	5	125
5	**6**	**77**	**1925**

The qualitative composition of the macroinvertebrates in the inflow of River Golema is comprised of 6 species from 6 systematic groups-one species from each group: Oligochaeta, Hirudinea, Gastropoda, Bivalvia, Amphipoda and Insecta. The highest density has been registered in the population of *Potamothrix hammoniensis*, 1250 Ind/m^2. In the samples, two species are endemic to Lake Prespa, while *Dreissena presbensis* is endemic to the Balkans. The presence of these species explains the faunistic exchange between the lake and its tributary. The value of the IBI is 2, and it indicates a poor ecological status, which despite the fact that it doesn't meet the WFD objective, is higher than the ecological status of river Golema in its middle course. The comparison chart (Fig.16.) shows the values of the density, diversity and IBI values. It is obvious that the results of the density and diversity are almost the same for both sampling seasons, while the IBI has a value of 2, which indicates a slightly improved ecological status, i.e. from bad to poor.

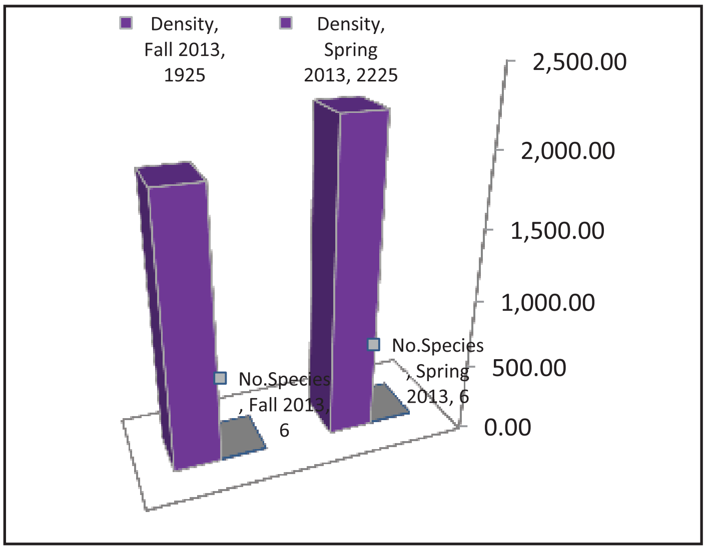

Figure 16. Comparison: density vs diversity in the Inflow of R. Golema (Spring vs Fall)

Table 15 shows the values of the traditional metrics. It is obvious that, based on these values, the ecological status is clearly bad for the Middle Course of River Golema in both seasons, while there are some variations concerning the values of the metrics for the sampling site River Golema Inflow. As it is shown in Table 15, the ecological status, depending of the metrics, varies in the boundaries between poor and bad. The poor structure (poor density and diversity) of the macroinvertebrates from the Middle Course of River Golema could have the influence of the assessment of the ecological status based on the traditional metrics because the reliability of these indexes increases with the number of specimens in the sample. Our results confirm the bad ecological status of River Golema which was previously assessed with the investigation in frame of the project Prespa Lake Watershed Management Plan (Prespa Lake Watershed Management Plan, 2013). In this study the macroinvertebrates were not found at all in some of the investigated sampling points of River Golema due to the excessive organic pollution.

Table 15. The ecological status of the sampling sites along River Golema based on the traditional metrics and the values of IBI index

River Golema	RG Inflow Spring	RG Inflow Fall	RG Middle Spring	RG Middle Fall
Number of individuals - N	2225	1925	2770	800
Number of taxa - S	6	6.0	2	2.0
Diversity Shannon & Weaner - H	1.52	1.7	0.9	1.0
Species reachnes po Margalef - d	2.56	1.5	0.29	0.3
Species evenness Pielou - e	0.59	0.7	0.9	1.0
Species dominance index Simpson - c	0.46	0.5	0.57	0.5
IBI-Irish Biotic Index	1	1	1	2

4.3. Lake Prespa Littoral

All of the four sampling sites in Lake Prespa are positioned in the northern part of the Lake: Oteshevo, NW of Ezerani, Ezerani and NE of Ezerani.

4.3.1. Oteshevo

4.3.1.1 Spring Sampling Campaign

The samples from this sampling site have been collected from 5 depth points, starting from 0.5-1 m to 5 m, which corresponded with the lowest distribution of the macrophyte vegetation (Tab.16.). Three types of habitats have been examined: sandy, muddy and muddy bottom covered with macrophyte vegetation. The general picture of both the density and the diversity of the macrozoobenthic communities are poor. The total number of species is 6, belonging to 5 classes: Hirudinea, Gastropoda, Bivalvia, Amphipoda and Insecta. The species composition indicates a certain level of pollution. The highest diversity has been recorded at 3-3.5 m and 5 m, on a muddy bottom and on a muddy bottom with a presence of macrophyte vegetation. The highest density has been reached among the population of endemic *Gammarus triacanthus prespensis*- 1200 Ind/m^2 at 1.5 m of depth on a sandy bottom with a presence of detritus. The highest density has been recorded (2425 Ind/m^2) on the bottom facies covered with vegetation and detritus at a depth of 3-3.5m.

Table 16. Qualitative and quantitative composition of the macroinvertebrates in Oteshevo, Lake Prespa in spring 2013

Locality: Otesevo Coordinates: 40°58'40.854"N; 20°54'59.738"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m^2	Total ind/m^2
0.5-1	Hirudinea	*Erpobdella octoculata*	3	125	
	Amphipoda	*Gammarus triacanthus prespensis*	6	400	525
1.5	Amphipoda	*Gammarus triacanthus prespensis*	6	2425	
	Bivalvia	*Dreissena presbensis*	4	100	2525
2.5	Gastropoda	*Valvata piscinalis*	3	100	
	Insecta	*Chironomus plumosus*	2	800	900
3-3.5	Amphipoda	*Gammarus triacanthus prespensis*	6	1200	
	Bivalvia	*Dreissena presbensis*	4	200	
	Gastropoda	*Valvata piscinalis*	3	125	
	Insecta	*Chironomus plumosus*	2	800	2325
5	Gastropoda	*Valvata piscinalis*	3	25	
		Pyrgula presbensis	3	25	
	Insecta	*Chironomus plumosus*	2	775	
	Amphipoda	*Gammarus triacanthus prespensis*	6	150	975
	5	**6**	**ASPT=3.8**	**7250**	**7250**

Among the registered species, there are two that are endemic to Lake Prespa: *Gammarus triacanthus prespensis* and *Pyrgohydrobia prespaensis* and one Balcan endemic species-*Dreissena presbensis*. The value of ASPT was 3.8 which indicates a bad ecological status.

4.3.1.2 Fall Sampling Campaign

Table 17. Qualitative and quantitative composition of the macroinvertebrates in Oteshevo, Lake Prespa in fall 2013.

Locality: Otesevo Coordinates: 40°58'40.854"N; 20°54'59.738"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
0.5-1	Oligochaeta	*Potamotrix hammoniensis*	2	50	
	Hirudinea	*Erpobdella octoculata*	3	25	
		Glossiphonia complanata	3	25	
	Bivalvia	*Dreissena presbensis*	4	750	
		Pisidium casertanum	6	25	
	Gastropoda	*Radix pinteri*	3	75	
		Gyraulus stankovici	3	50	
	Gastropoda	*Pyrgohydrobia prespaensis*	3	150	
		Bithynia prespensis	3	50	
	Insecta	*Baetis vernis*	4	25	1225
3	Oligochaeta	*Potamotrix hammoniensis*	2	25	
	Gastropoda	*Prespolitorea valvatiformis*	3	125	
	Bivalvia	*Dreissena presbensis*	4	1675	
	Insecta	*Chironomus plumosus*	2	25	1850
5	Oligochaeta	*Rhynchelmis komareki*	2	25	
		Pothamotrix hammoniensis	2	50	
	Gastropoda	*Prespolitorea valvatiformis*	3	75	
		Bithynia prespensis	3	25	175
	6	**13**	**ASPT=3**	**3250**	**3250**

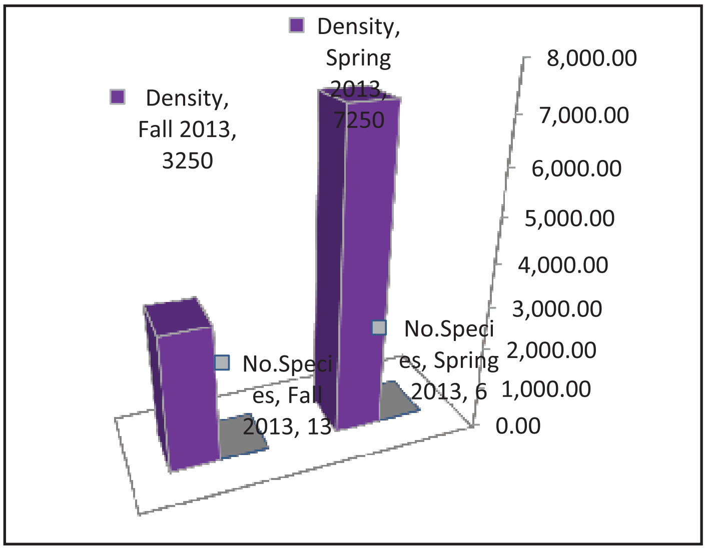

Figure 17. Comparison: density vs diversity in Oteshevo, Lake Prespa (Spring vs Fall)

13 species have been identified during the fall sampling campaign in Oteshevo sampling site (Tab.17.). They belong to 5 systematic groups: Oligochaeta, Hirudinea, Gastropoda, Bivalvia and Insecta. The highest diversity has been recorded at a depth of 0.5-1 m on a mostly stony bottom covered with macrophyte vegetation and with a presence of Dreissena dead shells. The highest individual (species)

density has been reached in the population of the Balcan endemic *Dreissena presbensis*-1675 Ind/m². 6 species (46 %) are endemic to Lake Prespa, including the Balcan endemic species *Dreissena presbensis*. The highest diversity (38 % of all species) has been registered in the class of Gastropoda. In Figure 17, the comparisons between the density and diversity in both seasons is shown. It is obvious that the density in spring is two times higher compared to the density in fall. The opposite happens regarding the diversity. In fall, the diversity is two times higher compared to spring. This phenomenon could be explained as follows: during the spring circulation of the water the food becomes an available resource which enables an optimum development of the density of the macroinvertebrates. During the summer, more species develop, the food becomes less available and this triggers a competition among the species, which becomes a limiting factor for the increase of the species population density. So, a bigger diversity, but a reduction in food availability influences the overall density which decreases during the fall period. The ASPT value was 2, which indicated a bad ecological status.

4.3.2. NW of Ezerani

4.3.2.1 Spring Sampling Campaign

Table 18. Qualitative and quantitative composition of the macroinvertebrates in NW of Ezerani, Lake Prespa in spring 2013.

Locality: Ezerani NW Coordinates: 40°59'45.26"N; 20°59'43.52"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
0.5-1	Oligochaeta	*Lumbriculus variegatus*	*2*	75	
	Hirudinea	*Erpobdella octoculata*	*3*	50	
	Amphipoda	*Gammarus triacanthus prespensis*	*6*	175	
	Gastropoda	*Planorbis prespensis*	*3*	50	
	Insecta	*Chironomus plumosus*	*2*	75	425
3	Oligochaeta	*Potamothrix hammoniensis*	*2*	125	
		Tubifex tubifex	*2*	200	
	Bivalvia	*Dreissena presbensis*	*4*	275	600
5	Bivalvia	*Dreissena presbensis*	*4*	650	
	Insecta	*Chironomus plumosus*	*2*	325	975
	6	**8**	**3**	**2000**	**2000**

Table 18 shows the macroinvertebrates density and diversity in the sampling site NW of Ezerani. The samples have been collected from three depth points (0.5-1; 3 and 5m). Three types of habitats have been a subject of research: sandy-muddy (mostly sandy); muddy-sandy (mostly muddy) and muddy habitat. The macrophyte vegetation has been present at 3m of depth, on a muddy sandy bottom, with a lot of empty shells from Gastropoda and Bivalvia (shell zone). The total number of species identified at the sampling site NW of Ezerani amounted to 8 species from 5 systematic groups: Oligochaeta, Hirudinea, Gasstropoda, Bivalvia, Amphipoda and Insecta.The highest diversity has been registered at the lowest depth of -0.5-1 m, on a bottom covered with sandy-muddy facies and with a presence of macrophyte vegetation. The highest species density has been reached at *Dreissena presbensis*, while the most density populated depth point was the depth point of 5 m, covered by muddy facies. The presence of Oligochaeta species, as well as Hirudinea and *Chironomus plumosus* from Chornomidae (Insecta) is indicative of water ecosystem under an anthropogenic impact, i.e. polluted ecosystems. Only one species from the list shown in Table 17 is endemic for Prespa Lake-*Planorbis prespensis*, while the Balcan endemic-*Dreissena presbensis* was present too, reaching the highest density among all registered species. The ASPT has a value of 3 and indicated a bad ecological status.

4.3.2.2 Fall Sampling Campaign

Table 19. Qualitative and quantitative composition of the macroinvertebrates in NW of Ezerani, Lake Prespa in fall 2013.

Locality: Ezerani NW Coordinates: 40°59'34.464"N; 21°01.0.732"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
1	Oligochaeta	*Rhynchelmis komareki*	2	125	
		Criodrilus lacuum	2	50	
		Limnodrilus hoffmeisteri	2	250	
	Amphipoda	*Gammarus triacanthus prespensis*	6	25	
	Gastropoda	*Prespolitorea valvatiformis*	3	25	475
3	Oligochaeta	*Limnodrilus hoffmeisteri*	2	50	
	Gastropoda	*Prespolitorea valvatiformis*	3	25	
	Insecta	*Chironomus plumosus*	2	25	100
5	Bivalvia	*Dreissena presbensis*	4	800	
	Insecta	*Chironomus plumosus*	2	200	1000
	5	7	**ASPT=2.8**	**1575**	**1575**

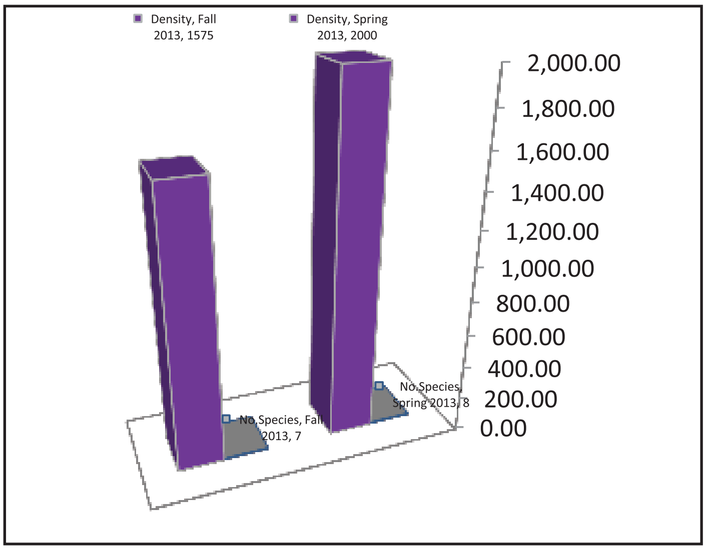

Figure 18. Comparison: density vs diversity in NW of Ezerani in Lake Prespa (Spring vs Fall)

The samples have been collected from three depth points. The community structure was poor in general: 7 species from 5 systematic groups have been identified (Tab.19.). The highest diversity was recorded at 0.5-1m, while the highest density per depth point was recorded at the depth of 5 m, on a mostly muddy bottom with a presence of many dead *Dreissena* shells. *Dreissena presbensis* reached the highest maximum-800 Ind/m². The presence of Oligochaeta (43 % of the total species number) and Chironomidae are indicative of polluted waters. The endemic species are less present compared with the sampling site Oteshevo. Both the density and the diversity differ only slightly in both seasons. The value of ASPT is 2.8, and it indicates a bad ecological status.

4.3.3. Ezerani

4.3.3.1 Spring Sampling Campaign

In Ezerani, the samples have been collected from 4 depth points: 0.5-1, 1.5-2, 3.5-4 and 5 m of depth. Two types of bottom facies have been considered as a factor impacting the macrozoobenthic communities' structure: sandy-muddy and muddy bottom. The bottom on the first depth points was sandy-muddy, while the other two were muddy bottoms covered with a macrophytic vegetation.

The biodiversity in Ezerani sampling sites comprises 7 species from 5 systematic groups. The highest diversity has been recorded in the class of Oligocheata-3 species (Tab.20.). In general the highest diversity has been recorded at a depth of 1.5-2 m, on the bottom with a sandy-muddy cover and with a presence of plant detritus-4 species.

Table 20. Qualitative and quantitative composition of the macroinvertebrates in Ezerani, Lake Prespa in spring 2013

Locality: Ezerani Coordinates: 40°59'36.98"N; 21° 0'25.63"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
0.5-1	Insecta	*Chironomus plumosus*	2	625	
	Oligochaeta	*Limnodrilus hoffmeisteri*	2	125	
	Hirudinea	*Erpobdella octoculata*	3	25	775
1.5-2	Gastropoda	*Valvata piscinalis*	3	25	
	Oligochaeta	*Eiseniella tetraedra*	2	25	
		Pothamotrix hammoniensis	2	75	
	Insecta	*Chironomus plumosus*	2	175	300
3.5-4	Gastropoda	*Valvata piscinalis*	3	50	
	Amphipoda	*Gammarus triacanthus prespensis*	6	75	
	Insecta	*Chironomus plumosus*	2	1475	1600
5	Insecta	*Chironomus plumosus*	2	800	800
	5	**7**	**ASPT=2.6**	**3475**	**3475**

The highest density was recorded at a depth of 4-4.5 m, on a muddy bottom covered with macrophyte vegetation. The highest species density has also been reached at a depth of 4-4.5 m, in the species *Chironomus plumosus* 1475 Ind/m². The highest density of *C. plumosus* which is an indicator of eutrophic waters, as well as the presence of Oligochaeta representatives typical for polluted waters, are indicative of a presence of pollution in this sampling point. The only endemic species registered at Ezerani was *Gammarus triacanthus prespensis*. The value of ASPT was 2.6 and it indicated a bad ecological status.

4.3.3.2 Fall Sampling Campaign

The table shows the qualitative and quantitative composition of the macroinvertebrates in the Ezerani sampling site during the fall. 7 species from 4 systematic groups have been identified. 43 % of the species are of the class of Oligochaeta. There are no endemic species in the Ezerani sampling site. The most densely populated depth point is at 0.5-1, where the bottom is covered with muddy facies with densely developed macrophyte vegetation. Here, the highest diversity has been recorded as well. The species *Limmnodrilus hoffmeisteri* has the highest density, while the Oligochaeta are distributed at all depth points.

Table 21. Qualitative and quantitative composition of the macroinvertebrates in Ezerani, Lake Prespa in fall 2013.

Locality: Ezerani Coordinates: 40°59'36.978"N; 21°00'25.625"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
1	Oligochaeta	*Rhynchelmis komareki*	*2*	75	
		Limnodrilus hoffmeisteri	*2*	250	
		Eiseniella tetraedra	*2*	25	
	Insecta	*Leptocoerus sp.*	*10*	25	375
3	Oligochaeta	*Limnodrilus hoffmeisteri*	*2*	25	
	Amphipoda	*Gammarus triacanthus prespensis*	*6*	25	
	Insecta	*Chironomus plumosus*	*2*	25	75
5	Oligochaeta	*Limnodrilus hoffmeisteri*	*2*	150	
		Eiseniella tetraedra	*2*	25	
	Amphipoda	*Gammarus triacanthus prespensis*	*6*	100	
	Gastropoda	*Bithynia sp.*	*3*	25	
	Insecta	*Chironomus plumosus*	*2*	50	350
	4	**7**	**ASPT=3.4**	**800**	**800**

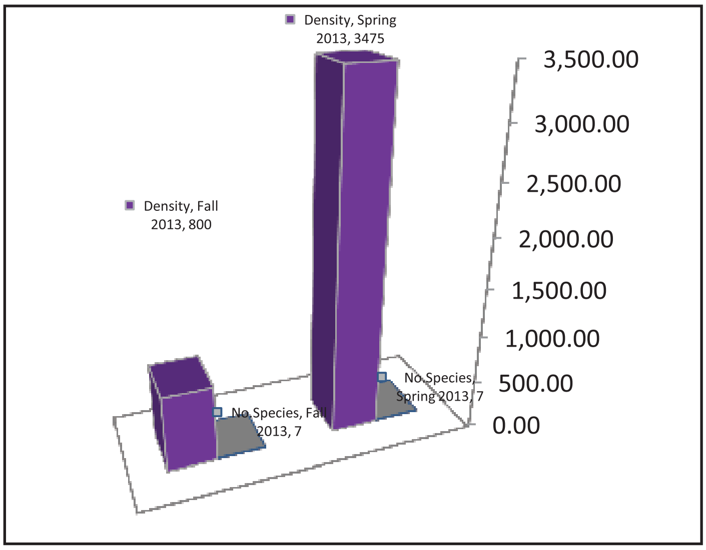

Figure 19. Comparison: density vs diversity in Ezerani, Lake Prespa (Spring vs Fall)

Fig.19 gives us the comparisons between the density and diversity in both seasons in the sampling site of Ezerani. The diversity is the same in both seasons while the density if far higher during the spring sampling season. The value of ASPT is 3.4, and it indicates a heavy polluted water or a bad ecological status.

4.3.4. NE of Ezerani

4.3.4.1 Spring Sampling Campaign

In NE of Ezerani, the samples have been taken from 3 depth points and three different facies: sandy-muddy (0.5-1m), mostly sandy (3m) and mostly muddy at 5m of depth. The macrophyte vegetation was very rare, and it is present at a depth of 0.5-1 and 5 m with some sparse *Najas sp.* species.

Table 22. Qualitative and quantitative composition of the macroinvertebrates in NE of Ezerani, Lake Prespa in spring 2013.

Locality: Ezerani NE Coordinates: 40°59'34.46"N; 21° 1'0.73"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m^2	Total ind/m^2
0.5-1	Gastropoda	*Limnaea pinteri*	3	50	
		Planorbis prespensis	3	50	
		Gyraulus stankovici	3	50	
	Oligochaeta	*Limnodrilus hoffmeisteri*	2	75	
	Decapoda	*Decapoda, Atydae*	5	100	325
3	Oligochaeta	*Limnodrilus hoffmeisteri*	2	50	
	Bivalvia	*Dreissena presbensis*	4	275	
	Gastropoda	*Prespolitorea valvatiformis*	3	25	
	Insecta	*Chironimus plumosus*	2	25	**375**
5	Oligochaeta	*Potamothrix hammoniensis*	2	50	
		Eiseniella tetraedra	2	50	
	Amphipoda	*Gammarus triacanthus prespensis*	6	50	
	Bivalvia	*Dreissena presbensis*	4	175	325
	7	**11**	**3.2**	**1025**	**1025**

The diversity at NE Ezerani is the highest among all the other 4 researched sites in Lake Prespa. 11 species have been identified belonging to 6 systematic groups: Oligochaeta, Gastropoda, Bivalvia, Amphipoda, Decapoda (Atyidae) and Insecta. The highest diversity-4 species have been recorded in the class of Gastropoda, 3 of which are endemic of Lake Prespa. Concerning the endemic species, beside the three mentioned above, the Balcan endemic species *Dreissena presbensis* has also been a part of the macroinvertebrates in this locality. The general picture of the macroinvertebrates in this locality is quite different compared to the other three localities. Here, , in a qualitative sense, the representatives from Gastropoda predominate, most of which are endemic forms. Additionally, although the species typical for eutrophic waters are present, their populations are quantitatively poor (*Chironomus plumosus* and *Limnodrilus hoffmeisteri).* All of this leads us to the conclusion that the water quality should be better compared to the other three sampling sites.

Concerning the total density per depth or habitat, it is noticeable that the density is more or less almost the same at all three depths. The same is true for the diversity, which is slightly higher at 0.5-1 m, on the bottom sandy-muddy bottom with a presence of macrophyte vegetation. Another specificity of this sampling site is the presence of the species from Decapoda, Atyidae which haven't been recently registered. The ASPT value was 3.2 and it indicates a bad ecological status.

4.3.4.2 *Fall Sampling Campaign*

Table 23. Qualitative and quantitative composition of the macroinvertebrates in NE of Ezerani, Lake Prespa in fall 2013.

Locality: Ezerani NE Coordinates: 40°59'45.258"N; 20°59'43.525"E					
Depth (m)	Class	Species (Total Number)	BMWP	Ind/m²	Total ind/m²
0.8-1	Oligochaeta	*Rhynchelmis kommareki*	2	50	
		Tubifex tubifex	2	25	
	Amphipoda	*Gammarus triacanthus prespensis*	6	50	
	Insecta	*Hermetia sp.*	5	50	175
3	Oligochaeta	*Rhynchelmis kommareki*	2	75	
		Tubifex tubifex	2	50	
		Eiseniella tetraedra	2	25	
	Bivalvia	*Dreissena presbensis*	4	300	
	Insecta	*Chironomus plumosus*	2	75	525
4-5	Oligochaeta	*Eiseniella tetraedra*	2	25	
	Hirudinea	*Erpobdella octoculata*	3	25	
	Amphipoda	*Gammarus triacanthus prespensis*	6	1075	
	Bivalvia	*Dreissena presbensis*	4	3000	
	Gastropoda	*Bithynia tentaculata*	3	25	
	Insecta	*Chironomus plumosus*	2	25	4175
	6	**9**	**ASPT=3.1**	**4875**	**4875**

9 species from 6 systematic groups have been identified at the sampling site NE of Ezerani during the fall sampling campaign. The highest diversity is present in the class of Oligochaeta-34 % of the species belong to this class. The highest density per depth point was recorded at the depth of 4-5 (Tab.23.) on a muddy bottom with a dense population of *Dreissena presbensis*. The highest density per species is also recorded at the depth of 4-5 m in the population of *Dreissena presbensis*. At the same depth highest diversity-6 species has been recorded.

The Figure 21 shows the comparison between the density and the diversity in spring and fall. Concerning the diversity, it is slightly higher in spring, while the density is much higher in fall. Concerning the density, it should be stressed that two species (*Dreissena presbensis* and *Gammarus triacanthus prespensis*) participate with more than 90 % in the density of the total macroinvertebrates.

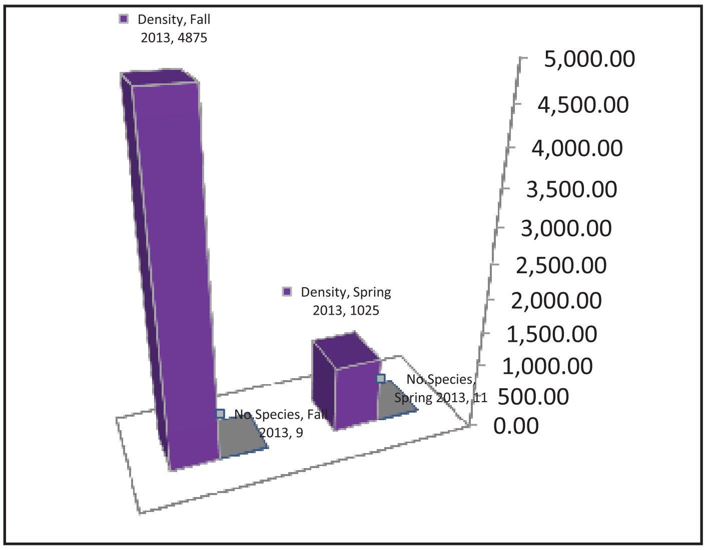

Figure 20. Comparison: density vs diversity in NE of Ezerani in Lake Prespa (Spring vs Fall)

The species composition (Tab.23.) differs between spring and fall. In the spring period, 4 endemic species have been registered while in fall, only two endemic species were registered. The representatives from Atyidae have not been registered during the fall period. These differences could be a result of the changes in the life conditions such as: general depletion of the food resources or variation (decreasing) of the water level due to the summer dry period. On the other hand, the increased density in some of the species could be explained either by the presence of species specific type of food resources that enable the population to develop to their maximum, which is in accordance with their physiological adaptation, or the fall season is the season which overlaps with the species maximum life cycle. The value of the ASPT is 3.1, and it indicates a bad ecological status.

The ecological status of the sampling sites in Prespa Lake has also been assessed using the traditional metric, unlike the rivers, where the Irish biotic Index was used.

Table 24 shows the colorful values for the indexes each of them representing certain ecological status.

Table 24. Values of the "traditional" metrics and ASPT values in the sampling sites in Lake Prespa in both seasons

Metrics	**Oteshevo**		**NW Ezerani**		**Ezerani**		**NE Ezerani**	
	Spring	Fall	**Spring**	Fall	**Spring**	Fall	**Spring**	Fall
Number of individuals - N	7250	3250	2000	1575	3475	800.0	1025	4875
Number of taxa - S	6	13.0	8	7.0	7	7.0	11	9.0
Diversity Shannon & Weaner - H	1.47	1.6	2.31	2.1	0.79	2.1	2.76	1.4
Species reachnes po Margalef - d	2.03	3.4	2.12	1.9	2.8	2.1	3.32	2.2
Species evenness Pielou - e	0.57	0.4	0.77	0.7	0.28	0.8	0.80	0.4
Species dominance index Simpson - c	0.44	0.6	0.28	0.3	0.79	0.3	0.23	0.5
ASPT	3.8	3	3	2.8	2.6	3.4	3.2	3.1

It is obvious, based on the colors, that almost all sites are characterized with an ecological status which is below "good", which is the objective of the WFD. The worst ecological status is in the sampling site of Ezerani, which was expected, having in mind the closeness of the Golema River which is a major polluter of the northern littoral of Lake Prespa, as well as the influence of the waste water treatment Plant in Ezerani. In the sampling site of Oteshevo, as it is shown in table 24, the ecological status varies between moderate and bad, depending of the index. In the locality NW of Ezerani, the ecological status varies between good and bad, while on the Ezerani sampling sites, according to all indices, the ecological status is bad. Slightly better conditions have been recorded at the locality of Ezerani NE where, the ecological status varies from high to bad. The bottom facies in Ezerani NE, which was mostly sandy-muddy with a presence of vegetation, could also have played a role for the higher diversity of the benthic fauna, compared with the other sampling sites where the bottom was mostly muddy, or more uniform. Also, the distance of the other three sites from Ezerani which is the most heavily polluted by the Waste Water Treatment Plant and the influence of the River Golema, could be considered as a factor why these three sampling sites were characterized with a better ecological status then the site of Ezerani. Here, especially in the most distant sampling points, such as Ezerani NE, the pollution effect has been mitigated in the process of auto purification. This fact, together with the heterogeneity of the bottom, could be the reasons for the better ecological status, based on the structure of the macroinvertebrates. Based on the values of ASPT, all sampling sites in Lake Prespa are classified as localities with bad ecological status.

5. CONCLUSIONS

5.1. Lake Ohrid Watershed-Tributaries

Using macronivertebrate community structure, i.e. metrics, an assessment of the ecological status of the sampled water bodies was done. Thus, it has been assessed, based on IBI values that the upper course of River Sateska is characterized with a high ecological status in both spring and fall. Going along the river from the upper course to the inflow, the ecological status deteriorates to poor both in spring and fall, with the exception of the inflow into the lake in fall, which is characterized by bad ecological status. All other sampling localities were assessed as having a bad ecological status. It has been already explained about the geomorphology and the differences in the anthropogenic impact on the different part of the rivers, which consequently leaded to changes in their ecological status. Hereafter we suggest delineating them (especially River Sateska) in two water bodies: Upper River Sateska, on one side and the Middle Course and its Inflow, on the other.

5.2. Lake Prespa Watershed-River Golema and Lake Prespa

A clearly bad ecological status has been registered at both sampling sites at River Golema, concerning the metrics of the macroinvertebrates community structure. The same ecological status has been determined for the remaining 4 sampling sites in the littoral part of Lake Prespa, meaning the sampling sites in the watershed of Lake Prespa and the Lake itself are at risk of failing to achieve the environmental objective of good ecological status according to the European Water Framework Directive. It is more than evident that the highest anthropogenic impact on the sampling sites in the littoral of Lake Prespa was identified in the northern part of the Lake, i.e. in the Ezerani localities (Ezerani, Ezerani NE and Ezerani NW). Going on southeast of the Lake, the pollution decrease which is reflected in the benthic communities' structure: here, there is a shift of the macroinvertebrates communities from Oligochata-Hirudinea, to Gastropoda, Bivalvia which are more sensitive to pollution (author's earlier investigations). That is why, we suggest to delineate Lake Prespa in two water bodies: Northern part of Lake Prespa and South Eastern part of Lake Prespa. River Golema, based on the results from our investigations should be delineated as a separate water body which has already been suggested by Prespa Lake Watershed Management Plan (Prespa Lake Watershed Management Plan, 2013), as River Golema 7-8, which corresponds with some of the sampling sites in our investigations.

6. RECOMMENDATIONS

Recommendations for future investigations on macroinvertebrates in the watershed of Lake Ohrid:

- Macroinvertebrate (macroinvertebrates) research, historically, is lacking continuity. Thus, establishing a long-term monitoring, which would inevitably include macroinvertebrates is imperative for a successful implementation of the European WFD.
- Having in mind the great depth of Lake Ohrid, where almost 2/3 of the bottom belongs to the profundal zone, the monitoring of the macrozoobenthic fauna should be extended from the littoral to the sublittoral and profundal region of the Lake.
- Developing a saprobic value system (indicator values) for the endemic species which predominate in most of the littoral, sublittoral and profundal part of the Lake should assist in overcoming the problem with the application of the specific and more accurate metrics which represent a base of the WISER system for assessment of the ecological status of the Lake.
- One of the premises of the European Water Framework Directive is the river basin management approach, which emphasizes the necessity of an extensive research on the tributaries in the watershed of the Lake.
- Global warming and invasive and alien species are a serious threat to the functionality of the Lake Ohrid ecosystem and should be given adequate consideration and further research should be undertaken in this field, too.

Recommendations for future investigations on the macroinvertebrates in the watershed of Lake Prespa:

- The benthic fauna (macroinvertebrates) researches, historically lack of continuity. Thus, establishing a long-term monitoring, which would inevitably include macroinvertebrates is imperative for the successful implementation of the Europan WFD.
- The monitoring of the macrozoobenthic fauna should be extended from the littoral to the sublittoral and profundal region of the Lake.
- Developing a saprobic value system (indicator values) for the endemic species which are a constitutive part of the macrozoobenthic communities of Lake Prespa would assist in overcoming the problem with the application of the specific and more accurate metrics which represent a base of the WISER system for assessment of the ecological status of the Lake.
- One of the premises of the European water frame directive is the Integrative Basin River management approach, which emphasizes the necessity of extensive research on the tributaries in the watershed of the Lake.
- To get a more realistic picture of the ecological status of the Lake, more sampling sites should be distributed in different parts of the Lake respecting the criteria of the differences in the anthropogenic influence and hydrogeomorphology of the Lake.
- Global warming and invasive and alien species are a serious threats to the functionality of the Lake Prespa ecosystem and should be given an adequate consideration and Intense research should be undertaken in this field, too.

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Annex 1.

Table 25. A summarized preview of the macroinvertebrates results in the tributaries of Ohrid and Prespa lakes.

	LAKE OHRID TRIBUTARIES										LAKE PRESPA TRIBUTARIE			
	River Sateska						River Koselska		River Cherava		River Golema			
Species/Season	Upper Course		Middle Course		Inflow		Inflow		Inflow		Middle Course		Inflow	
	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall	Spring	Fall
Species Abundance (ind/m^2)	5150	6050	2875	12075	3725	10550	800	925	1850	775	2775	800	2225	1925
OLIGOCHAETA														
Criodrilus lacuum		1												
Potamothrix hammoniensis		1		2				8					14	50
Limnodrilus hoffmeisteri							1					17	14	
Stylodrilus sp.							7							
HIRUDINEA														
Glossiphonia complanata	1				1									
Erpobdella octoculata			4	4	3		2	3					1	3
Haemopis sanguisuga									2					
GASTROPODA														
Ancylus fluviatilis	1	2		1										
Theodoxus fluviatilis			3											
Lymnaea stagnalis								3						
Planorbis prespensis													2	2
BIVALVIA														
Dreissena presbensis														8
Pisidium casertanum													1	
AMPHIPODA														
Gammarus balcanicus	36	47		27										
Gammarus roeseli			64	170	83	375				7				
Gammarus ochridense										8				
Gammarus triacanthus prespensis														9
ISOPODA														
Asellus aquaticus								1						
INSECTA														
EPHEMEROPTERA														
Ephemera danica	17	33		94	38	29	1			7				
Baetis vernus	32	39	4	30			8			4				
Baetis scambus				7										
Baetis rhodani				3				3						
Epeorus pleuralis	4	8												
Ecdyonurus venosus	4	5		14	3									
Rhitrogena sp.	23													
Ephemerella sp.	6													
Leptophlebia sp.						6								
Caenis macrura							1							
PLECOPTERA														
Perla marginata	14	21												
Isoperla sp.	1	3		3										
Leuctra ferruginea	1			2	1									
TRICHOPTERA														
Limnephilus sp.	1		4	2	3									
Hydropsyche sp.	1													
Leptocerus sp.	49	49		111	12	1								
Silo sp.	3													
Sericostoma sp.	6	16			2	3		1		5				
Goeridae				2		1								
Rhyacophila sp.				1										
DIPTERA														
Tabanus sp.	1		1	4										

Atheryx sp.	3		3											
Chironomus plumosus		13		3		4		18			76	15	57	5
Chironomus sp.							10		64					
Tanypus sp.	1		24								35			
Bezzia sp.	1													
Tipula sp.		4							2					
Ormosia sp.				1			1							
Hermetia sp.							1							
ODONATA														
Gomphus vulgatissimus			2		1	2								
Aeshna grandis									1					
Coenagrion sp.									1					
COLEOPTERA														
Limnius sp.			4	1	1									
Dytiscus sp.				1										
Platambus maculatus									4					
HETEROPTERA														
Aphelocherius aestivalis			2		1									
Corixa sp.						1								
Total Abundance	206	242	115	483	149	422	32	37	74	31	111	32	89	77
No. of taxa/species	21	14	11	21	12	9	9	7	7	5	2	2	6	6
Shannon W. Diversity	3.3	3.1	2.15	2.7	1.93	0.7	2.5	2.1	2.3	2.3	0.9	1	1.52	1.7
Margalef Species Richness	8.64	5.5	4.85	7.5	5.06	3	5.3	3.8	3.7	2.7	0.29	0.3	2.56	1.5
Pielou Eveness	0.75	0.8	0.62	0.6	0.54	0.2	0.8	0.8	0.9	1	0.9	1	0.59	0.7
Simpson Dominance Index	0.14	0.1	0.36	0.2	0.38	0.8	0.2	0.3	0.25	0.2	0.57	0.5	0.46	0.5
Irish Biotic Index (IBI)	5	5	3	4	3	2	1.5	2	2	2	1	2	1	1

Legend: The colors are in accordance to the colorful scale for ecological status in frame of the European WFD, where each color represents different ecological status:

- Red=Bad ecological status
- Orange=Poor ecological status
- Yellow=Moderate ecological status
- Green=Good ecological status
- Blue=High ecological status

Annex 2.

Table 26. A summarized preview of the macroinvertebrates results in the littoral part of Lake Prespa

Lake Prespa Sampling Sites	Oteshevo								Ezerani NW								Ezerani								Ezerani NE							
	Spring				Fall				Spring				Fall				Spring				Fall				Spring				Fall			
	Abundance by maximum depth (m) and total abundace (T)																															
	1	3	5	T	1	3	5	T	1	3	5	T	1	3	5	T	1	3	5	T	1	3	5	T	1	3	5	T	1	3	5	T
Species Abundance (Ind/m²)	3050	3225	975	7250	1225	1850	175	3250	425	600	975	2000	475	100	1000	1575	1075	1600	800	3475	375	75	350	800	325	375	1500	2200	175	525	4175	4875
OLIGOCHAETA																																
Rhynchelmis komareki							1						5								3								2	3		
Potamothrix hammoniensis					2	1	2			5							3										2					
Lumbriculus variegatus									3																							
Tubifex tubifex										8																			1	2		
Limnodrilus hoffmeisteri													10	2			5				10	1	6		3	2						
Eiseniella tetraedra																	1				1		1				2			1	1	
Criodrilus lacuum													2																			
HIRUDINEA																																
Glossiphonia complanata					1																											
Erpobdella octoculata	5				1				2								1														1	
GASTROPODA																																
Valvata piscinalis		9	1														1	2														
Pyrgohydrobia prespaensis			1		6																											
Radix pinteri					3																											
Gyraulus stankovici					2																				2							
Bithynia prespensis					2		1																									
Bithynia tentaculata																															1	
Bithynia sp																							1									
Planorbis prespensis									2																2							
Prespolitorea valvataeformis						5	3						1	1												1						
Limnea pinteri																									2							

BIVALVIA		8																														
Dreissena presbensis	4				30	67				11	26				32											11	7			12	120	
Pisidium casertanum					1																											
AMPHIPODA																																
Gammarus triacanthus prespensis	113	48	6						7				1					3				1	4				2		2		43	
ISOPODA																																
Asellus aquaticus																																
DECAPODA																																
Atyidae																									4							
INSECTA																																
EPHEMEROPTERA																																
Baetis vernis					1																											
Leptocerus sp.																					1											
DIPTERA																																
Chironomus plumosus		64	31			1			3		13			1	8		32	59	32			1	2			1				3	1	
Hermetia sp.																													2			
Total Abundance	122	129	39	290	49	74	7	130	17	24	39	80	19	4	40	63	43	64	32	139	15	3	14	32	13	15	60	88	7	21	167	195
No. of taxa/species			6				13				8				7				7				7				11				9	
Shannon W. Diversity			1.47				1.6				2.31				2.1				0.79				2.1				2.76				1.4	
Margalef Species Richness			2.03				3.4				2.12				1.9				2.8				2.1				3.32				2.2	
Pielou Eveness			0.57				0.4				0.77				0.7				0.28				0.8				0.80				0.4	
Simpson Dominance Index			0.44				0.6				0.28				0.3				0.79				0.3				0.23				0.5	
ASPT-Average Score Per Taxon			3.8				3				3				2.8				2.6				3.4				3.2				3.1	

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Legend: The colors are in accordance to the colorful scale for ecological status in frame of the European WFD, where each color represents different ecological status:

- Red=Bad ecological status
- Orange=Poor ecological status
- Yellow=Moderate ecological status
- Green=Good ecological status
- Blue=High ecological status

"HYDRA"
OJF

Fish Monitoring and Fisheries

Lake Ohrid

Final Report

Lead authors:

Zoran Spirkovski – HIO

Dusica Ilik-Boeva – HIO

Co-authors:

Ekerem Kapedani – HYDRA

Arian Palluqi – HYDRA

Aleksandar Flloko – HYDRA

Trajce Talevski – HIO

Technicians:

Tom Miraku & Bardhyl Radovicka - HYDRA

Gjorgi Puftoski & Zaim Ramadan - HIO

Student volunteer: Blagoja Trajcevski -HIO

Contents

1. Summary

The Project Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar regarding fish and fisheries component gave in some cases improved knowledge about Lake Ohrid fish stock. In this case, the sampling campaigns in the autumn period gives main contribution for the ecological moments of the alien species correlated with the habitats, and regarding the main commercially valued fish species very little knowledge improvement was achieved.

The Project Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar regarding Lake Ohrid fish fauna gives a good exercise of possibilities to evaluate the implementation of EN 14757 MMG. But, in such large water body using sampling procedure based on stratified random sampling of fish assemblages out of their aggregation periods as spawning and winter schooling can give only partial knowledge about ecological status of fish population and the ecological status of the water body.

From 135 randomly set MMGN in the 2013 sampling campaign 16 fish species were detected in total from which only two alien species, stone moroko - *Pseudorasbora parva* and bitterling – *Rhodeus amarus*. The most abundant species at certain sub-basins are: at SB1 Veli Dab is spirlin – *Alburnoides hridanus* with 37%, at SB2 Andon Dukov is the roach - *Rutilus ohridanus* with 40%, at SB3 Radozda is stone moroko with 29%, at SB4 Central plate is Ohrid gudgeon –*Gobio ohridanus* with 100%, at SB5 Lin is the bleak – *Alburnus scoranza* with 22%, at SB6 Hudunisht is the roach with 27% and at SB7 Tushemisht is the roach with 44%. Except for the roach which is dominant at three sub-basins, in all other sub-basins different species are predominant.

Regarding the number of individuals at all sub-basins overall dominance has the roach progressively towards the depth whilst the spirlin or the bleak are the second dominant native species changing from one to other sub-basin and mainly in the depth strata till 20m. As fourth native significant from the biodiversity point native species present in number of individuals is the Ohrid minnow – *Pachychilon pictum,* or the Ohrid gudgeon – *Gobio ohridanus* changing from one to other sub-basin. Distribution of species expressed in biomass per strata follows more or less the case of number of individuals in most of the sub-basins. For SB7 significant presence of rudd – *Scardinius knezevici* and chub – *Squalius squalus* in the biomass of catch is the main difference from all other sub-basins. From the commercially important species during this sampling campaign no specimen of Ohrid trout was caught and only 27 individuals of Ohrid belvica – *Salmo ohridana* and 5 individuals of carp – *Cyprinus carpio* were caught.

Other significant moment is that during this sampling campaign no specimens of Ohrid nase - *Chondrostoma ohridanus* wasn't caught at all. Form other hand, remarkable thing is the presence of the barbell – *Barbus rebeli* in almost all of the surveyed sub basins.

The composition of the species length (age) classes distribution is in the following manner: the bleak was present at all sub-basins except for SB2 and SB4 with quite different number of length classes from 3 at SB3 and SB7 till 12 at SB1, the spirlin is present at all littoral sub-basins with 4 classes at SB2 and 8-13 length classes at the rest of the sub-basins, the stone loach is present only at the three littoral sub-basins on Macedonian side and on the other side, the minnow (Phoxinus lumaireul) is present only at the three Albanian sub-basins. The presence of the endemic Ohrid minnow – Pachyhilon – with 12 and 13 length classes in the SB1, SB5 and SB6 shows in a manner the similarities of these water sub basins as habitats.

As it appears that the most dominant as in number, as well as in length classes the roach dominates again in this manner.

The whole thing regarding transboundary fish monitoring implementing the EN 14757 standard with only MMG gives positive benefits for the respectful institutional collaboration of the three countries sharing the three lakes and additional involvement of colleagues from other countries brought other ways of future perception of the fishery component within the EU WFD concept.

Regarding the last one, at Lake Ohrid, according the achieved results we could state that no drastic criteria can be used for classification of the water sub basins, like it is more expressed at Lake Prespa. But, compared also from the data gathered by personal communication with fishers from both countries, it is obvious that at the last several years more fish is gathering at the Albanian part of the lake. This mainly reflects to the smaller sized fish like bleak, roach, spirlin that are eurytopic fish with great tolerance in the ecological niches, but they also prefer more eutrophic conditions. This can be used like a valid index of habitat's situations regarding the water quality state of these sub basins.

This kind of situation can be result of reducing the nutrient and anthropogenic pollution load at the Macedonian part from one side by the operation of the waste water collecting system along the east to northwest part of the coast, and also the increment of naval recreational speed boats that are affecting the fish welfare.

Comparison of all other data in the classification of different parts of Lake Ohrid water and habitat's quality and state can perhaps raise the slight differences registered between the sub basins investigated by the fish status in them.

The results from this project, regarding the fish and fishery point, gives a good base to improve the further fish monitoring of the large Balkan lakes and especially Lake Ohrid due to its big depth with an average of 165m. This understands increment of fishing efforts, sampling campaigns in different time periods of the year, usage of other fish survey gears and technics and increasing the collaboration between the experts and actual fishing companies or FMOs.

2. Introduction

The aim of the CSBL project is to improve the implementation of legislation, regulations and management plans for the conservation and sustainable use of biodiversity at lakes Prespa, Ohrid and Shkodra/Skadar. Component 2 of the CSBL plan of operation is targeting on fish monitoring and fisheries. Main tasks within this component are the implementation of joint monitoring for Lakes Prespa, Ohrid and Shkodra/Skadar, human, institutional and organizational capacity building.

This report concerns only the results, preliminary conclusions and recommendations on the fish stock stage at Lake Ohrid – based on the fish monitoring by multi mesh size fishing standards and existing fishery statistics.

The agreed monitoring scheme is based on multi-mesh gillnetting according to the European Standard EN 147 57, as a central and common sampling element in all lakes. This kind of sampling is practiced for the first time on Lake Ohrid.

These requirements of the WFD in relation to fish communities are in assessing population status from species composition, abundance and age classes which in this case can contribute to the assessment of the current ecological state of Lake Ohrid.

3. Fish fauna and fisheries at Lake Ohrid

Lake Ohrid is a 290 m deep oligotrophic lake, shared between Macedonia and Albania, oldest in Europe (age of several million years) and with tectonic origin. It has catchment area of over 2.600 km^2 and surface of 358 km^2. Lake Ohrid has four permanent tributaries: Cerava, Velgoska, Koselska,Sateska and more than 30 temporary creeks and numerous underwater (sublacustrine) springs. It has a surface outlet, River Drim/Drin which belongs to the Adriatic drainage system .

Three cities are situated around the lake, two in Macedonia, Ohrid and Struga, and Pogradec in Albania with in total 150.000 inhabitants which are including also more than 10 villages.. Tourism, small and medium enterprises are main source of income for the majority of the citizens and there is no intensive industry or agriculture in the region in the last three decades.

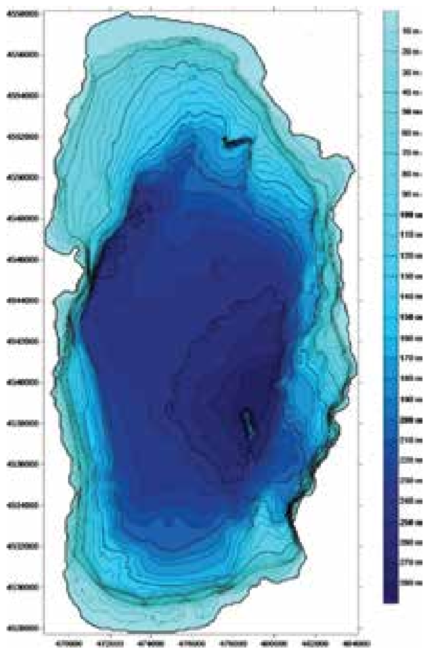


Figure 1. Bathymetric map of Lake Ohrid
(Source: Wilke, T.**, Väinölä, R. & Riedel, F. (eds.) (2008)**

Lake Ohrid is UNESCO natural world heritage site from 1980 and the eastern and south-eastern part of the lake shore as well as the spring area in St. Naum are under protection of biodiversity in Galicica National Park in Macedonia.

Lake Ohrid is a subtropical oligomictic lake and by both physic-chemical parameters and phytoplankton, according EU WFD is an oligotrophic lake. There are no anoxic layers in the water column. Even in the deepest part of the lake oxygen level never drops below 6 mg/l. The water is exceptionally clear with transparency to a depth of as much as 22 meters. It has been estimated (Stankovic 1960), that the retention time of the lake water volume is 83 years. Being carstic basin of Lake Ohrid (Hoffmann et all 2012), it shows still in nowadays tectonic activities, which from other hand are renewing the water volume (bottom sinking) and improving the "self-purification") ability of this ecosystem.

The biggest water volume is oligotrophic (low primary productivity) and though poor in fish yield also.

3.1. Fish fauna and stocks

Lake contains a lot of endemic species, with nearest neighbours in Pliocene. The size of Lake Ohrid and the quality of the fish fauna, gives to the lake an important significance from the fishery point. The quality and the economic value of the fish populations of this lake are on a much higher level than the rest of the Balkan's lakes, even above those shallow and high productive ones from the Aegean zone (Stankovic, 1960). The fish fauna is represented with 17 autohthonous species from four families: Salmonidae (2), Cyprinidae (12), Cobitidae (2) and Anguillidae (1) and 6 alochthonous species or in total twenty-one taxa. None of them is migratory species.

Table 1. Lake Ohrid fish species (native and alien)

Species Latin name	Species common name	native species	alien species (year of introduction)
Alburnoides ohridanus	Ohrid spirlin	+	
Alburnus scoranza	Bleak	+	
Anguila anguila	European eel	+	
Barbatula sturanyi	Stone loach	+	
Barbus rebeli	Barbell	+	
Carassius gibelio	Prussian carp		+ (1983)
Chondrostoma ohridanus	Ohrid nase	+	
Cobitis ohridana	Spined loach	+	
Cyprinus carpio	Carp	+	
Gambusia holbrooki	Mosquito fish		+ (1940's)
Gobio ohridanus	Ohrid gudgeon	+	
Lepomis gibbosus	Pumpkinseed		+ (1990's)
Oncorhynchus mykiss	Rainbow trout		+ (1974)
Pachychilon pictum		+	
Pelasgus minutus	Ohrid minnow	+	
Phoxinus lumaireul	Minnow	+	
Pseudorasbora parva	Stone moroko		+ (1970's)
Rhodeus amarus	Bitterling		+ (1990's)
Rutilus ohridanus	Ohrid roach	+	
Salmo ohridana	Ohrid belvica	+	
Salmo letnica	Ohrid trout		
Squalius squalus	Ohrid chub	+	

In the fishery 10 species have commercial value with prior to the two relic and endemic trout - *Salmo letnica* (Karaman) and *Salmo ohridana* (Steind) - than the European eel *Anguilla anguilla* (L.) and the bleak *Alburnus scoranza* (Filipi). Also there are several temporary present alohthonous species, among which only *Carassius gibelio* (Bloch) is evident in the fishery catch. According to the previous fishery statistic at the Macedonian part of the lake, for the periods 1930/57 (Stankovic, 1960), 1929/73 (Tocko, 1975), the salmonid fishes and the eel were represented with 45.6% of the mean annual catch where 43% belongs only to *Salmo letnica*. Due to this fact, from the fishery aspect the lake itself was characterised like a typical salmon lake. The same statistics were used to estimate the mean annual fish yield per unit of lake's surface and it has a value of cca 9 kg/ha, for the Macedonian part. This in other hand again shows the scarcity of nutrients in the lake and in the same time its oligotrophic character. All these things make the fishery as an important economy branch in this part of the country.

Besides its scientific and economic value Lake Ohrid's ichthyofauna hasn't been so much investigated in manner of presence of alien species and mainly those recordings have been represented sporadically without any continuous monitoring attention.

As in other fishery intensive exploited water bodies most of the work has been paid to commercially valuable species mainly Lake Ohrid trout, Lake Ohrid belvica, eel, carp and bleak.

The ichthyologic investigations were mainly addressed to reproduction of the native species their forage and the relation between cyprinid and salmonid species in the terms of their food competitiveness (zooplanktophagues).

From the data in certain technical report or papers provided for the fishery sector of both countries Albania and Macedonia changes of the assemblages of the bleak particularly of their winter schooling are quite evident. Normally in front of the villages Trpejca, Pestani, Radozda on Macedonia side and Lin and Memlisht on the Albanian part of the Lake the bleak was schooling till 2003, after that things were changed – they were not schooling at this places but they were spread all over the lake as in littoral as well in pelagic part.

From other technical and research papers changes in the spawning ecology of the two endemic Lake Ohrid trouts were evidenced and recorded due to habitat disturbance.

Before the Second World War and after the War in the combat against the malaria mosquito fish was introduced into Lake Ohrid.

In the beginning of the 60's of the past century at the River Drim (Drini) forming like an outlet of Lake Ohrid and entering the Adriatic See, on the Macedonian side two dams were constructed for electric hydropower production and later on, on the Albanian side three more were constructed. Concerning the fishery this had let to cut of the natural recruitment of the eel population in the lake. From other hand enlarging of the watershed of the Lake Ohrid with additional 463 km^2 with the diversion of the River Sateska which was naturally a tributary to River Drim, large area of trout spawning grounds on the north part of the Lake were devastated due to erosion (siltation) and nutrient load.

In 1974 the presence of rainbow trout was for the first time recorded in the lake and examples could be found regularly in the fish catch until 1994. Its presence was result of existing rainbow trout farm on the Albanian side, close to the shore, which was closed with join Albanian-Macedonian decision in 1994. This fish farm was converted to a hatchery and nursery of the Lake Ohrid trout fingerlings.

In 1983 evidence of Prussian carp like a new present species in the Lake Ohrid fish fauna was recorded, so in certain years the catch of this species exceeds more than 20 tons annually.

In second half of the 90's bitterling was introduced accidently during the transport of the silver carp stocking material for fish farms in Albania. Until now there isn't recorded evidence of symbiotic relations between bitterling and lake shells. In the same decade and most likely together with the bitterling the sunfish was introduced.

At present drastic decline of Lake Ohrid trout is still evident which started from the beginning of 90's. Both Lake Ohrid trout and Lake Ohrid belvica due the overfishing and still high fishing pressure are drastically endangered even on the edge of their population irreversibility. Despite all the efforts from both countries sharing the lake of improving the lake Ohrid trout with the joint restocking programme, the results cannot be visible due to fishing the in mature specimens, and in this case manly by the poachers. The bleak population is in expansion, eel population is maintained with stocking material, carp population shows relative good conditions. It's worth to mention manly n that almost 2,5 decades the population of undermouth (nase) has been also drastically declined as in the catch as well in previously known habitats. Similar situation was recorded with the barbell population. The rest of the endemic but commercially unvalued species are in a relatively good condition, depending on temporal changes in their habitats.

Regarding the alien species rarely specimens of rainbow trout can be found still in the fish catch. The same situation is with the silver carp. The rest of alien species which are minnows are still present with different abundance on different habitats, but their impact on the rest of the fish fauna and the rest of the ecosystem hasn't been investigated yet.

Detailed inventarisation and also fish stock assessment was never performed on Lake Ohrid fish. Despite all the efforts of the scientists and technical people involved in fishery of Lake Ohrid from both countries to attract, implement and raise this issue on a higher level which it deserves till now no funds have been allocated for this manner - a real fishery investigation.

In table 1 two trout species are presented *Salmo letnica* (Lake Ohrid trout) and *Salmo ohridana* (Lake Ohrid belvica) despite the five explained in the literature (Kottelat and Freyhof 2007), since never in the fishery of Lake Ohrid four forms of *Salmo letnica* (*Salmo balkanicus*, *Salmo typicus*, *Salmo aestivalis* or *aphelios* and *Salmo lumi*) were treated as separate species.

Bellow in a section for fisheries is given historic data for dominant species and stock estimates of species of economic importance.

3.2. Fisheries

Due to the long period of transition from one to the other political systems both in Albania and Macedonia, fishery was also conveyed several times to different levels of control and organization.

Albania

During the previous regime (before 90's), the fishery activity in Albanian part of lake Ohrid was managed by State Enterprise and there were about 35 fishers. After the regime collapsed, there were about 800 fishers (legal and illegal). The legal fishers were organized in different groups or cooperatives. By 2002, all licensed fishermen were organized in a Fisheries Management Organization which has responsibility and duty to manage a landing site, and to participate in the co-management of fisheries resources. The FMO has a limited number of licenses (110).

Fisheries Management Organization relating to the Relevant Co-management Area is involved in the preparation and implementation of the Co-management Plan.

Co-management Plan lasts for a maximum period of ten years and aim:

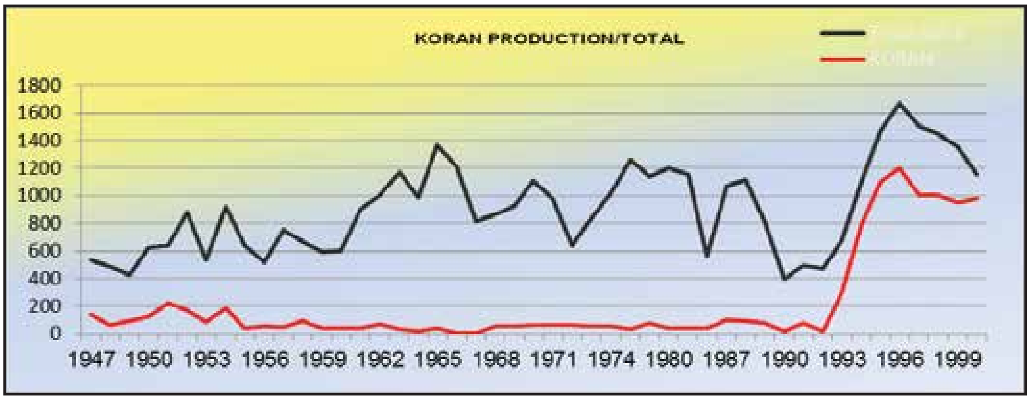
a) to promote the utilization of fishery resources based on the sustainable development;

b) to maintain the quality and biological diversity of fisheries resources;

c) to encourage the use of appropriate fisheries technology; and

d) to avoid the creation of excess fishing capacity

The FMO, based on implementation of the law "On Fishery and Aquaculture" and its specific regulations, have to apply management measures.



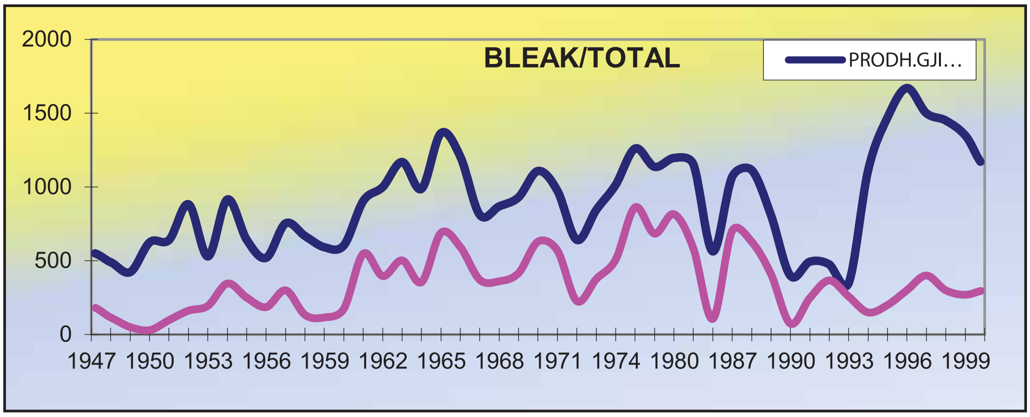


Figure 1. Total catch, trout and bleak catch in the period 1947-2000 (Source: MoEFWA – Fishery)

Catch statistics for the Lake Ohrid (1947-1993)

Due to the centralized regime of the period all data in the table are taken from (taken from governmental data).

- The increase of belushka (*Salmo ohridana*) and bleak (*Alburnus scoranza*) catch is due to the use of bottom trawl and purse seine fishery from 1962-1990.
- Squalius catch includes also the following species (Barbus – approx. 20.5 %), (Gobio – approx. 1.5%) and (*Scardinus* – approx. 0.5 %).
- In the period 1947-1988 (*Rutilus*) was included in the bleak statistics representing nearly 12-15% of the bleak catch data.
- In the period 1947-1993, fishery was a state activity.
- During the 1947-1993 period 0.5-1 % of the trout catch was represented by the *Salmo letnica forms Salmo lumi* and *Salmo balcanicus.*

The drastic fall of undermouth after 1985, shows the poor water quality of the effluents, posing a problem for the spawning of the species into these streams.

Table 2. Total commercial annual catch for the period 1947 till 1993 in quintals (100 kg)

Description	1947	1950	1955	1960	1965	1970	1975	1980	1985	1986	1987	1988	1989	1990	1991	1992	1993
Lake Ohrid trout	10	10	3	3,8	9,3	6,1	3	3,8	37	3,6	10	9,2	8	1,6	7,7	1,6	2,2
Lake Ohrid Belvica	4	3,2	1	12,5	30,1	11,6	12	9	9	7,8	11,7	12	10,5	6,6	1,7	1,3	1,2
Carp	8	26,9	11,7	4	8,7	2,4	2,9	11,9	45	3,5	4,5	17,3	11,5	12,5	5,7	4,4	1,5
Undermouth (nase)	6,5	10	8,4	7	8,4	10	13	4,5	1	0,9	0,3	1,3	0,3	0,2	0,4	0,1	0,2
Chub	7,3	10,4	15	13,6	11	17,7	8,8	6	10	9,9	10	8,7	4,7	6	4,8	0,5	0,8
bleak	17,7	3	25	17,5	68,9	63	86,2	81,6	58,5	10,6	70,3	63	41	7,4	24,7	36,7	21,8
roach													4,5	5	4,2	2,9	2,8
Total Catch	53,5	62,5	64,1	60,4	136,4	110,8	126	116,8	86,7	34,3	106,8	111,5	80,9	39,8	49,3	47,5	34,7

(Source: MoEFWA – Fishery)

The above data are official ones from the fishery directorate of Albania, whilst for the period after 1993 the data aren't reliable and are estimated like approximation, and they are shown in further Fig. 7 and 8.

According to some estimation, annual fish catch of 220 - 260 ton is reported during last years in which 75-80% of it is bleak, 5-10% Lake Ohrid trout and Lake Ohrid belvica. The rest belongs to roach and barbell.

Macedonia

In the period from 1945 to 1994 commercial and recreational fishery were organized and performed by two state enterprises, each responsible for one part of the lake because Macedonian part of the lake was divided into fishery sub basins Ohrid and Struga. These enterprises were employing professional fishers – licenced by the state issued from the fishery exams in the Hydrobiological Institute as Authorised institution for fisheries. Total number of professional fishers on Macedonian part of the lake was from 80-120 altogether.

Apart from this, two citizens associations like recreational fishers were existing and for the recreational fishery they were buying daily fishing licences from the fishery enterprises. The main object of recreational fishery was Lake Ohrid trout, and the fishing was performed by boats (4-5 m in length) using nylon ropes with spinner hooks and its worth to be mentioned that more than 2000 daily licences were issued per day during 15 days of recreational fishing. The fishing per person was limited to 4 specimens of Lake Ohrid trout per day, but that limit was highly exceeded and the catch from the recreational fishing per year was in some years represented by half of commercial fishing catch. These numbers were never part of the fishery statistics. In 1994 both state fishery enterprises were privatized (FAO – Macedonia Country report, 2005).

The fluctuations of the total annual catch from commercial fishery are presented on the following figures.

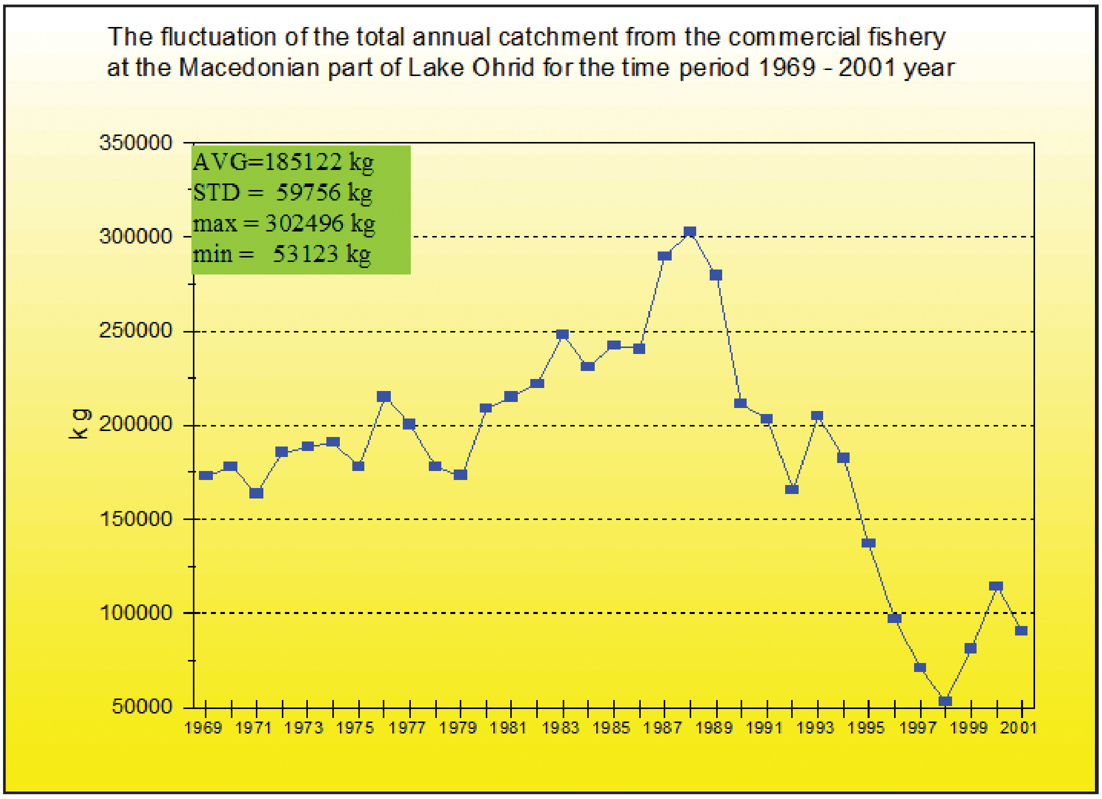


Figure 2. Total annual fish catch in the period 1969-2001 at the Macedonian part of the Lake Ohrid (Source: Spirkovski at all., 2002)

Differences in the fish catch between the first half and the end of the XX century and first years of this century is presenting shift from salmonid to cyprinid fish species.

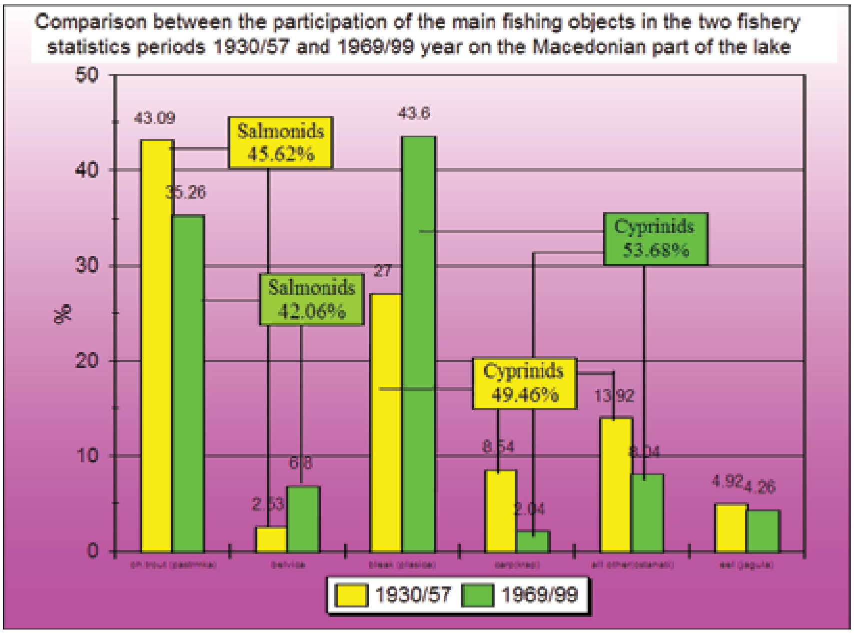


Figure 3. Comparison between two fishery statistics periods 1930/1957 and 1969/1999 for Lake Ohrid (Source: Spirkovski at all., 2002)

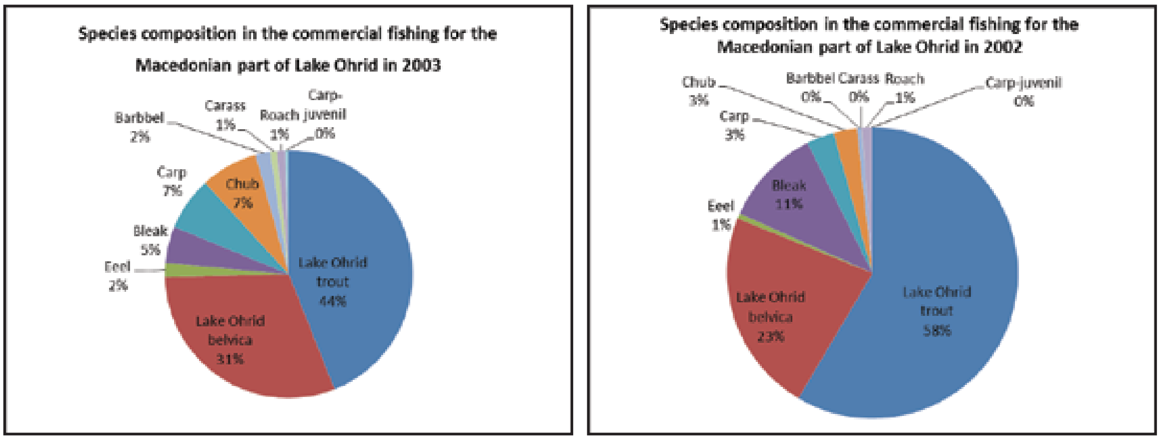


Figure 4. Species composition of the total annual fish catch in 2002 and 2003 at the Macedonian part of the Lake Ohrid (Source: Fisheries statistics AD Ohridska Pastrmka – Fishing company Macedonia)

Related to the overfishing and unsustainable fishery in April 2004 concession on the fish stock from Lake Ohrid was withdrawn by the state government and until September 2012 total moratorium on fishing was engaged. During this period only fishing for scientific purposes for artificial spawning of Lake Ohrid trout was performed.

After 8, 5 years of total moratorium on fishery at Lake Ohrid in September 2012 new Concession was tendered and signed. Currently 47 fishers are employed by the Concessionaire. On the following graph is presented species composition in the total catch in the period of the new concession.

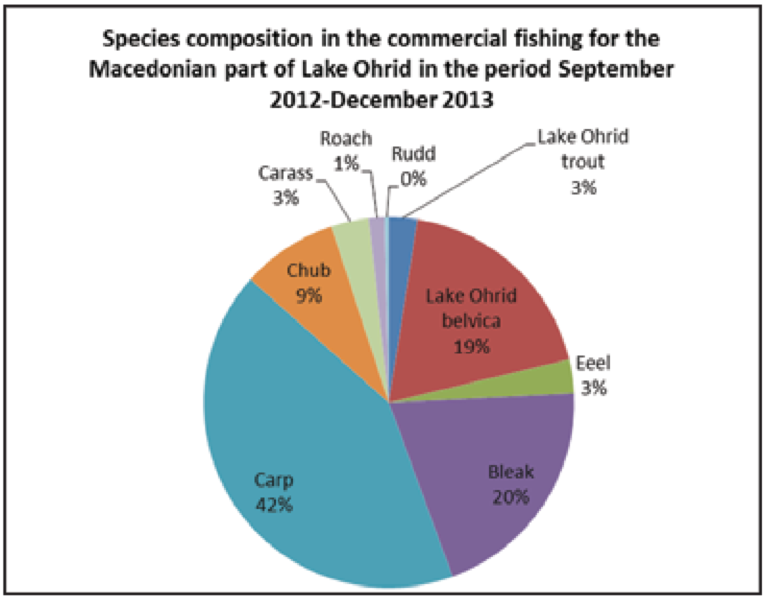


Figure 5. Species composition of the total fish catch in the period September 2012 – December 2013 at the Macedonian part of the Lake Ohrid (Source: Fisheries statistics Pastrmka 2012 Ohrid– Fishing company Macedonia

Figure 5 represents the period of adapting to fishing of the concessionaire company in terms of training fishermen, learning fishing habitats and as well relatively short fishing period.

3.2.1. Albania – Macedonia (Synthesis for Lake Ohrid)

From 1960 until 1990 Joint Fishery Commission Albania – Macedonia regulated quotas per country per species, restocking (Albania mostly for carp and Lake Ohrid trout and Macedonia for Lake Ohrid trout and eel), minimum allowed catchable sizes per species, closed seasons, etc. After the 1990 collaboration between two countries continued on a scientific level and on ministerial level but not official.

According to fisheries laws in both countries the fishing season is closed during spawning of all species, apart from regulated fishing activities to supply the hatcheries with sexual products for artificial breeding of Lake Ohrid trout.

Although 11 species of fish are comprising commercially, the most dominant ones are the Lake Ohrid trout, Lake Ohrid belvica, eel, carp, and bleak. The bleak has the lowest commercial value. Because of the predatory interactions and competition for food, when the trout abundance is high, bleak abundance is lower, and vice versa. This pattern is reflected in the catch statistics.

In Macedonia, the fishery statistics are based on the actual weight of the fish landed by commercial fishermen. In Albania, data before 1991 are based on weights landed, but after that date, landings are estimates only. These data clearly show that there has been a significant decline in total catch in the last decade of XX century (Fig. 6).

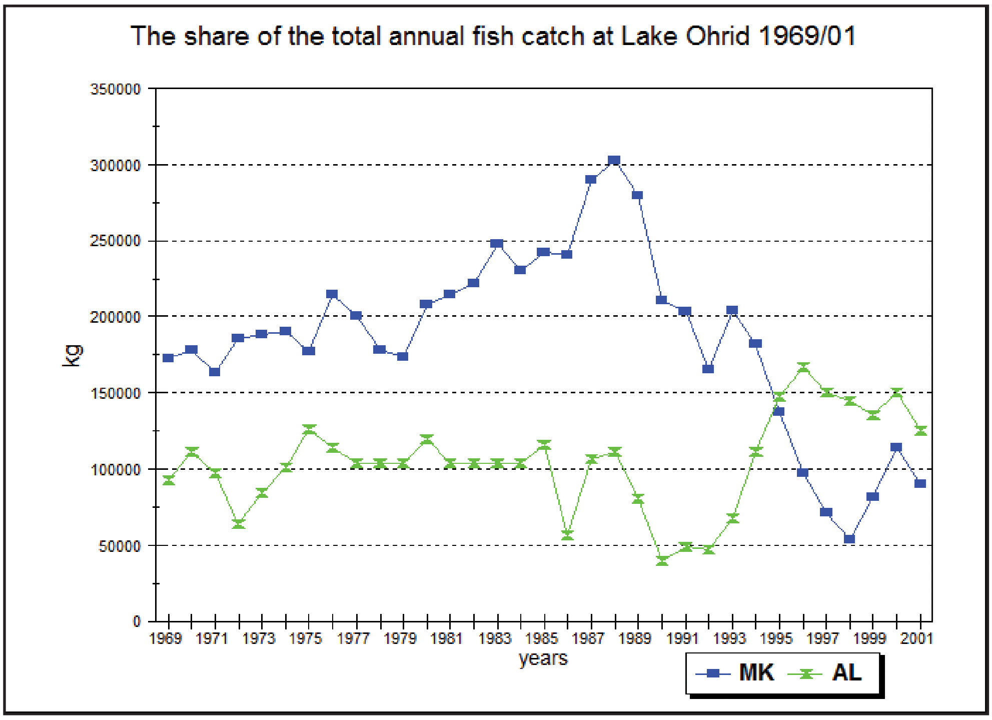


Figure 6. Total annual commercial fish catch in Lake Ohrid, by country (Source: Spirkovski at all., 2002)

Beginning in 1992, the landings in Albania increased dramatically, while those in Macedonia began to fall. This is especially apparent in the trout catch (Fig. 7).

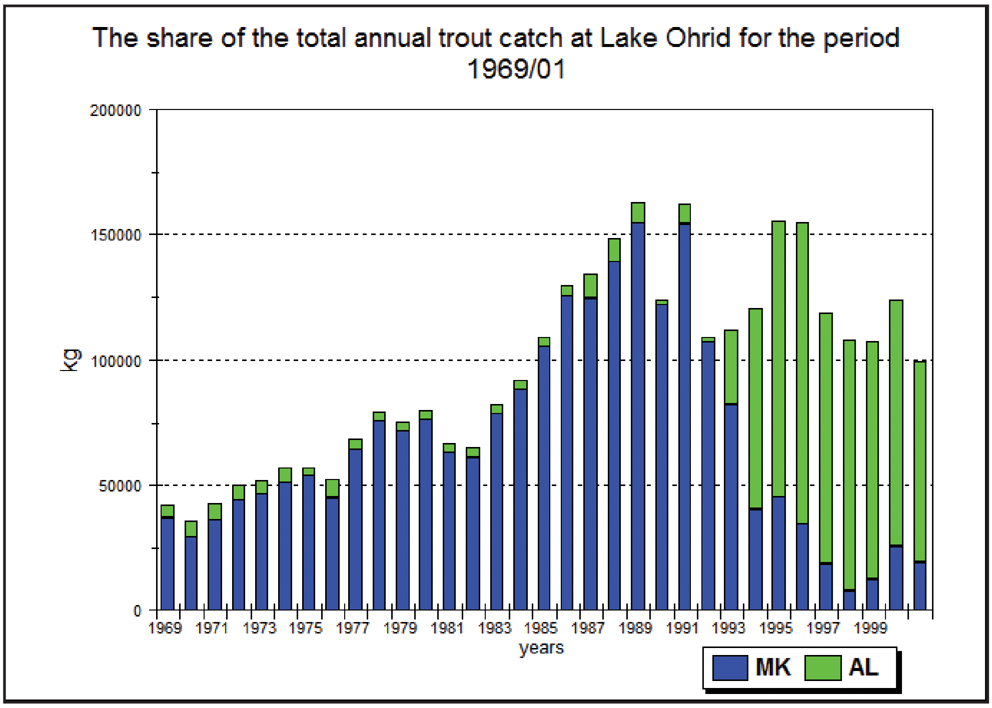


Figure 7. Annual trout catch in Lake Ohrid, by country (Source: Spirkovski at all., 2002)

The species composition of the catch has also changed substantially, with pressures on the trout populations increasing from 1969 to 2001 (Fig. 8). In the period (1999-2001) the trout made of about 48% of the total catch.

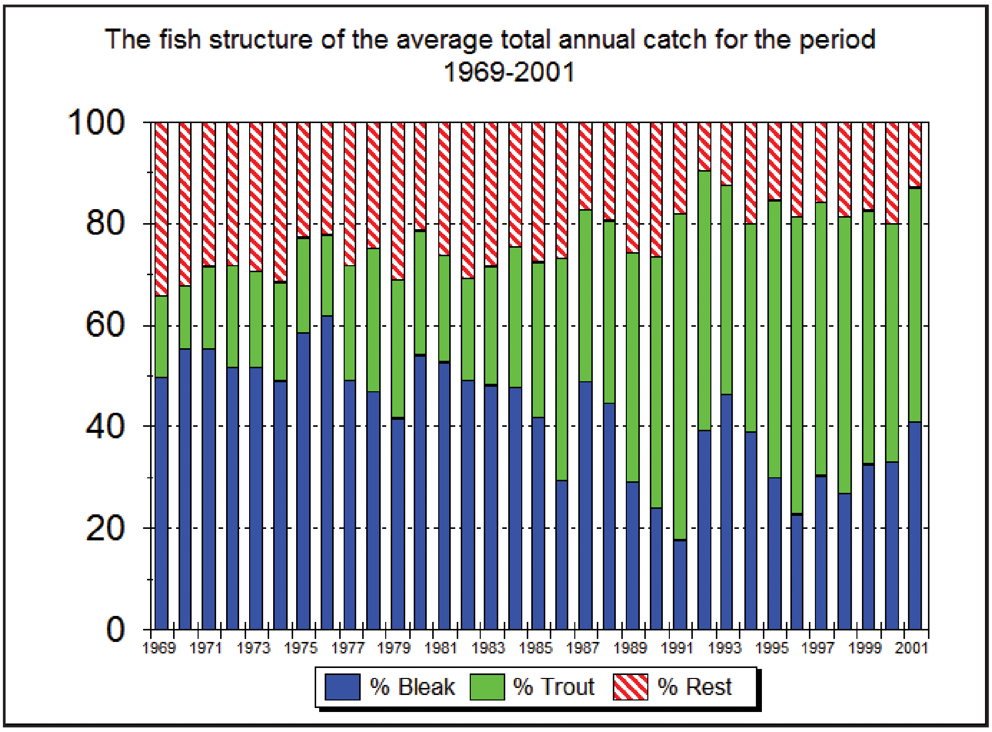


Figure 8. The species composition of the annual catch from Lake Ohrid. (Source: Spirkovski at all., 2002)

3.3. Restocking

According to fisheries laws in both countries the fishing season is closed during spawning of Lake Ohrid trout, apart from regulated fishing activities to supply the hatcheries with sexual products for artificial breeding.

Over the years various types of restocking material have been used simultaneously:

- **fry**
- **alevins of 95 days since fertilization**
- **fingerlings of six months after hatching**
- **young fish of 12 months old**

The success of stocking strategies remains unknown, but it is a fact that every spawning season fish are caught of spawning size in 50 to 60 mm meshed nets (including the fish that are still maturing and those which have already released their gonad products – naturally spawned).

Since 2005 at Macedonian side and 2009 at Albanian side all adult fish caught for artificial spawning process are released into the Lake waters after striping the gonad products so they can contribute in the reproduction in future years. For decades before this part of the reproductive population was ending on the market as fish for consumption.

On the following graph is presented restocking of Lake Ohrid with offspring of Lake Ohrid trout in different ontogenetic stages at Macedonian part.

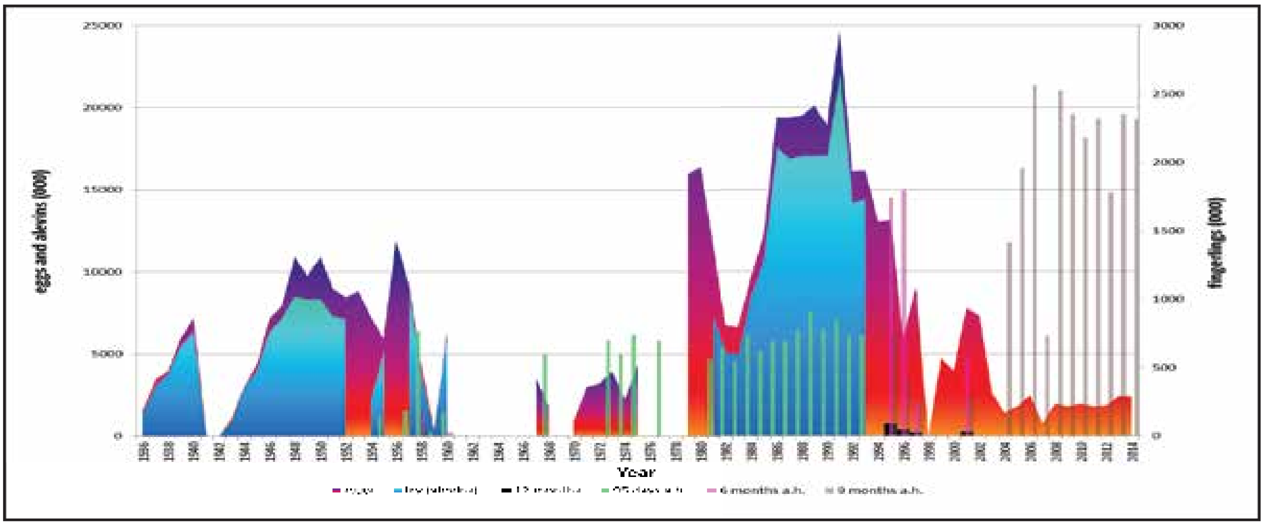


Figure 9. Lake Ohrid trout restocking on Macedonian side of the lake (Source: HIO Statistics)

Restocking at Albanian side started in the second half of the 60's in the XX century with Lake Ohrid trout fries (alevins) with approximately 250-300 thousand individuals per year until 2003 and 300 thousand fingerlings of carp with average weight of 5 g annually started in 1980 which stopped in 2005. From 2003 restocking in both countries is performed during the autumn zooplankton peak in the lake only with fingerlings of Lake Ohrid trout, nine months from fertilization and six months fed. On Albanian side is done with 750-900 thousand individuals. Due to the uncertainty regarding the success of trout stocking it is recommended to conduct marking and tagging experiments and to sample catches for hatchery-reared trout.

3.4. Legislation

Albania

The legislation framework in the fishery sector, in an overall overview is complete and contemporaneous. The legislation deals not only with fishery issues but also with other related issues such as: biodiversity, socio-economic aspects etc.

There are several legislative acts regulating the fishing activity, including:

The new **Law No. 64/2012** set the basis for the good management of the fishery sector explains many of the terms and concepts related to the fishery sector

It should be stressed that the main intention of the law are:

to ensure a rational and accountable exploitation of aquatic biological resources and development of aquaculture;

provide protective conservation measures in order to ensure the protection of biological water resources, and

support the sustainable development of fishery and aquaculture sectors, as well as create better socio - economic conditions for producers.

Relevant laws:

Law "On the Land" (1991)
Law "On Forests and Forestry Police" (1992)
Law "On Protection of Wild Fauna and Hunting" (1994)
Law "On Fishing and Aquatic Life" (1995)
Law "On Water Reserves" (1996)
Law "On the Regulatory Framework of the Water Supply Sector and of Disposal and Treatment of Waste Water (1996)

Law "On Environment Protection" (2011)
Law "On Protected Areas" (2002)
Law "On Protection of Marine Environment from Pollution and Damage" (2002)
Law "On Protection of Trans-border Lakes" (2003)
Law "On Environmental Impact Assessment" (2011)

Macedonia

In 2007 the existing Law on Fishery (from 1993) has been replaced with the Law on Fishery and Aquaculture (LFA) Official gazette7/2008 date15.01.2008. This law has three amendments: one in 2010 Official gazette 67/2010 date 14.05.2010 and two in 2011: Official gazette 47/2011 date 08.04.2011 and 53/2011 date 14.04.2011.

The following documents are complimentary to the Law on Fishery and Aquaculture:

Law for the protection of Ohrid, Prespa and Dojran Lake Official gazette 45/1977 date 09.09.1977. This law has four amendments: one in 1980 Official gazette 08/1980, one in 1988 Official gazette 51/1988 and one in 1990, Official gazette 10/1990 and one in 1993, Official gazette 62/1993.

Law for nature protection Official gazette 67/2004 date 04.10.2004 This law has five amendments: one in 2006 Official gazette 14/2006, one in 2007 Official gazette 84/2007, one in 2010 Official gazette 35/2010,and two in 2011 Official gazette 47/2011 and Official gazette 148/2011.

Law for the environment Official gazette 53/2005 date 05.07.2005, This law has seven amendments: one in 2005 Official gazette 81/2005, one in 2007 Official gazette 24/2007, one in 2008 Official gazette 159/2008, one in 2009 Official gazette 83/2009, two in 2010 Official gazette 48/2010 and 124/2010 and one in 2011 Official gazette 51/2011.

Fishery Master Plan for Lake Ohrid for the period 2011-2016. Official gazette 145/2011

and

Amendments of the Fishery Master Plan for Lake Ohrid for the period 2011-2016. Official gazette 57/2013

Regulations:

Regulation on the form, content and the way of performing evidence of fish production as for the amount of the sold fish per species 2008

Regulation for performing the fish guarding service, the form and the content of the fish guardian legitimation, as the way of its issuing and withdrawing 2008

Regulation of the content of the Program for examining, the form and content of the certificate, as the cost for issuing certificate for commercial fishery 2008

Regulation on the form and the content of the evidence formulary in the fishing regions 2008

Regulation of the content of the Fishery Master Plan 2008

Regulation of the content of the annual plan for protection and exploitation of the fish and the content of the annual report of realization of the plan 2008

Regulation on the technical requirements for the lending sites 2008

Regulation on the quality, size and weight, as also the way of declaring the fish for traffic market 2008

Regulation on the way of marking of the boats and tagging and evidencing of the fishing gear 2008.

Regulation on the form and the content of the document for the origin of the fish and the way of its issuing and fulfilling 2010

Regulation on the way of issuing licenses for recreational fishing, the required documentation for issuing, the form and content of the evidence formulary, the way of evidencing and delivering the data 2010

Regulation on the form and the content of the legitimation for recreational fishing and the way of its issuing 2010

Regulation on the allowed fishing gears and equipment and their use for commercial and recreational fishing 2011

Regulation on the length of the fish under which they cannot be fished for commercial and recreational fishing 2011

Regulation on the quality, size and weight, as also the way of declaring the fish for traffic market 2013

Regulation for amendments of regulation on the allowed fishing gears and equipment and their use for commercial and recreational fishing 2013

Regulation for changes of the on the length of the fish under which they cannot be fished for commercial and recreational fishing 2013

3.4.1. Comparative review of fishing/fishery rules in AL and MK

Fishing ban season per species for Macedonian part of Lake Ohrid is in 30 days in the spawning period which can differ from year to year, but has to be in the stated period in the following table.

Table 3. Fishing ban season by species and by countries

Common name	Latin name	ALBANIA		MACEDONIA	
Ohrid trout	*Salmo letnica*	1st December	28th February	15th October	31st March
Ohrid belvica	*Salmo ohridana*	1st November	31st January	15th October	31st March
carp	*Cyprinus carpio*	20th May	15th June	1st April	30th June
chub	*Squalius squalus*			1st May	31th May
nase	*Chondrostoma ohridanus*			1st April	31st May
rudd	*Scardinius knezevici*			1st April	31st May
barbell	*Barbus rebeli*			1st May	30th June
bleak	*Alburnus scoranza*	20th April	15th June	1st May	30th June
minnow-pachychilon	*Pachychilon pictum*			1st May	30th June

Table 4. Minimum allowed dimensions for some of the commercial species at Lake Ohrid

Common name	Latin name	ALBANIA	MACEDONIA
Ohrid trout	*Salmo letnica*	32 cm	35 cm
Ohrid belvica	*Salmo ohridana*	30 cm	22 cm
carp	*Cyprinus carpio*	30 cm	40 cm
chub	*Squalius squalus*	15 cm	25 cm
roach	*Rutilus prespensis*	12 cm	15 cm
bleak	*Alburnus scoranza*	10 cm	12 cm
rudd	*Scardinius knezevici*	12 cm	20 cm
eel	*Anguila anguila*	25 cm	60 cm
prussian carp	*Carassius gibelio*	15 cm	unlimited
pumpkinseed	*Lepomis gibbosus*		unlimited

4. Transboundary sampling scheme performed

4.1. Methodology of sampling

Multimesh gillnetting

The EU standard EN 14757 was used to design the modified sampling scheme according to Lake Ohrid specifications – large and deep water body, different substrates (habitats). In order to satisfy statistical model we avoid periods of fish grouping (spawn and wintering shoaling).

MMGN at the Macedonian part of the lake were used 10 of them with mesh size panels as described in the standard, but before usage previously we labelled each net with white cotton fabric with the number of the net from 1-10 and also each mesh panel with its dimensions for easier determination of the fish caught.

Between the actual realised and previous preliminary fish monitoring sampling scheme at the Macedonian side of the lake, the main deviations are in missing usage of different gears (all except benthic MMGN), because they were not succeeded to be delivered during the CSBL Project. Analogue to it the time schedule was not used foreseen for the gears of subject.

In the localities or sub basins as we named them afterwards, there isn't change, but according the joint Fishery TWG we expand the area around the proposed localities and subsequently renamed Daljan to Andon Dukov SB2, Kalishta to Radozda SB3 and introducing new sub basin like a replacement for the place where pelagic nets on 240 m depth had to be used, to SB4 but with MMGN, in order to have at least some results for the bottom fish fauna in October-November at this area – which lies between SB2 and SB3.

Figure 10. Lake Ohrid sampling sub basins (SB1-SB7)

The same applies to the Albanian part as to the planned and realised gear and periods, and also the change in the locality name; there was only referring to Pojske but extending like SB6 it was renamed to Hudunisht.

Having in mind the lake's characteristics, bathymetry, habitat differentiation, and previous long term practise in experimental fishing, we divided the whole lake in 7 sub basins (SB 1-7) which corresponding to their habitat constituency equally per country. SB4 appear like a new one from the previous design of the delineation of the sub water bodies, due to impossibility of using pelagic net at the deepest point of the lake.

Hence, following the Coriolis water current in the lake, we had the presented numerations (see the map).

The two teams (AL and MK) tried to choose most similar SB's which were corresponding as in habitat substrate, bathymetric configuration, wind exposures, and total ecological condition. (Annex I)

Three sub basins at Albanian and four sub basins at Macedonian side of the lake were sampled from which six are littoral and one pelagic sub basin.

Table 5. Lake Ohrid sampling sub basins

Country	Sub basin (SB)	Locality	Position
Macedonia	**SB1**	Veli Dab	littoral
	SB2	Andon Dukov	littoral
	SB3	Radozda	littoral
	SB4	Central plate	pelagic
Albania	**SB5**	Lin (Bakalicë)	littoral
	SB6	Hudenisht	littoral
	SB7	Tushemisht	littoral

Reciprocally SB1 (Veli Dab) and SB5 (Lin – Bakalicë) are localities with the following habitat distribution:

a. From the lake shore up to 6 meters there is a solid substrate, with gravel and stones of different dimensions that are predominant, and between them areas of sand and fine detritus. At the lake shore there are some areas with *Phragmites and Scirpus,* and some *Potamogeton*, and *Myriophyllum* further from the shore.

b. In 20 meters depth there is a muddy substratum with little sand and Charophyta vegetation.

c. In 20-50 meters depth there is a sandy area with mollusk's shell deposits.

d. Over 50 meters the substrate is sandy and with detritus.

Both localities Veli Dab and Lin-Bakalicë are intensive spawning areas for *Salmo letnica*, and the slope of lake's bottom in these two localities is steepest than in all other areas of the lake's littoral and sub littoral.

As for the previous two sub basins, SB2 (Andon Dukov) and SB7 (Tushemisht) are sub basins with similar habitat distribution and ecological conditions:

a. From the lake shore up to 6 m, there is fine substrate (mixture of sand and mud) and with presence of plant origin detritus.

b. From 6-20 m depth, there is a muddy area with intensive presence of meadows of Charophyta vegetation.

c. In 20-50 meters depth there is a sandy area with mollusk's shell deposits (although less than in Lin area).

d. Over 50 meters the substrate is sandy with detritus.

The following two sub basins, SB3 (Radozda) and SB6 (Hudenisht) are areas with these habitat descriptions:

a. From the lake shore up to 6 meters it is a sandy substrate with small size or stones and gravel. The vegetation is well distributed.

b. In 20 meters depth there is a muddy substratum with little sand and Charophyta vegetation.

c. In 20-50 meters depth there is a mostly sandy area with mollusk's shell deposits.

d. Over 50 meters the substrate is sandy and with detritus.

The SB4 (Central plate) is a substitution of previously planned sub basin for multimesh pelagic nets sampling, and at this sub basin sampling was performed at deeper strata with benthic multimesh nets.

Fish population sampling was done according to the European Standard CEN 14757 – standardised protocol, using benthic multi-mesh gillnets which are 30 m long and 1.5 m deep, composed of 12 panels with different mesh sizes ranging from 5 mm to 55 mm from knot to knot in the following order: 43mm, 19.5 mm, 6.25 mm, 10 mm, 55 mm, 8 mm, 12.5 mm, 24 mm, 15.5 mm, 5 mm, 35 mm and 29 mm.

Lake Ohrid sampling period started at 19.10.2013 following the previously agreed protocol, but due to the weather conditions and problems with the delivery of the fishing gears, sampling was postponed for two weeks and proceeds on 4th of November and finished on 12.11.2013.

Nets were set before the dusk, stayed overnight and after the down were taken out (12 hours of sampling) to cover both highest activity circadian peaks.

Nets were processed panel per panel, per species and measuring of the individuals was performed afterwards.

4.1.1. Albania

Setting the nets on more than one sub basin per night derived with organization scheme of two groups composed of 4 people (two fishermen and two HYDRA experts - one scientist and one technician). The different number of nets per night varied (8-16) and was depending on weather condition in different sub basin. Fishermen boats were used for setting and lifting the nets.

Figure11. Lake Ohrid fish sampling (AL)

Table 6. Summary table of multimesh gillnets sampling at sub-basins: Lin-Bakalice (SB5), Hudenisht (SB6) and Tushemisht (SB7)

WATER BODY	SUB BASIN Sampling date	No. of nets per sub basin	Maximum ind./net	Minimum ind./net	Nets / strata	
LAKE OHRID	LIN – Bakalice (SB5) (06.11.2013/08.11.2013/ 09.11.2013/13.11.2013)	24	189	32	0-3 m	6
					3-6 m	3
					6-12 m	3
					12-20 m	5
					21-35 m	2
					>35 m	5
	HUDENISHT (SB6) (05.11.2013/06.11.2013/ 13.11.2013)	24	169	17	0-3 m	4
					3-6 m	1
					6-12 m	4
					12-20 m	8
					21-35 m	3
					>35 m	4
	TUSHEMISHT (SB7) (08.11.2013/09.11.2013/ 12.11.2013)	24	73	5	0-3 m	4
					3-6 m	3
					6-12 m	3
					12-20 m	2
					21-35 m	5
					35-50 m	7

4.1.2. Macedonia

Sampling procedure was based on stratified random sampling. Depending on weather conditions, 8-10 benthic multi-mesh gillnets per night were set before dusk and were taken out after the down. Only testing of the pelagic nets was performed, but due to the high buoyance of those nets, for this sampling period they were not used. All pelagic nets were modified during the project and can be used now according to the standard in future samplings.

Figure 12. Pelagic nets testing

GPS coordinates for each net, net setting depth, setting position to the shore, Air and Water temperature, pH, Conductivity, O_2, transparency, Sechi disk depth and weather conditions were registered (Annex II). All nets per strata following the randomization scheme were set in different directions related to the shoreline. So, for example in one particular strata, some nets were set from the shore starting with the panel of 43 mm, some nets ending closer to the shore with the panel of 29 mm in some cases perpendicular to the shore in some cases in an angle of 45° or 60°, in other cases parallel with the shoreline.

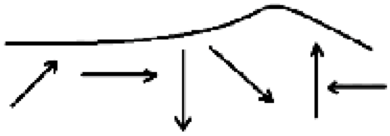


Figure 13. Random sampling

The research vessel of PSI Hydrobiological Institute was used for setting and lifting the nets in all sub basins of Lake Ohrid.

Figure 14 and 15. Lake Ohrid fish sampling

Table 7. Summary table of multimesh gillnets sampling at three sub basins: Veli Dab (SB1), Andon Dukov (SB2), Radozda (SB3) and Central plate (SB4)

WATER BODY	SUB BASIN	No. of nets per sub basin	Maximum ind./net	Minimum ind./net	Nets / strata	
LAKE OHRID	VELI DAB (SB1) (19.10.2013/04.11.2013)	16	560	10	0-3 m	2
					3-6 m	4
					6-12 m	4
					12-20 m	3
					21-35 m	3
	ANDON DUKOV (SB2) (08.11.2014/11.11.2014)	20	188	0	0-3 m	2
					3-6 m	3
					6-12 m	4
					12-20 m	4
					21-35 m	3
					35-50 m	3
					50-75 m	1
	RADOZDA (SB3) (05.11.2014/06.11.2014)	17	338	1	0-3 m	3
					3-6 m	4
					6-12 m	3
					12-20 m	2
					21-35 m	3
					35-50 m	2
	CENTRAL PLATE (SB4) (12.11.2014)	10	7	0	120-125 m	4
					130-135 m	3
					165 m	3
	Pelagic net	1	33		0-6 m	1

Considering how many individuals per species per panel were caught, in those sub basins where there were not more than 50 individuals, all caught specimens were measured. In cases where several hundreds of one species were caught per panel, 50 individuals were measured by length and weight and the total weight and number of individuals of the rest was recorded. Weight was measured on a portable balance with accuracy of 0.1 g. Standard and total length were measured to the closest mm and for data processing just total length was used and averaged to the closest cm. In total 7479 individuals were caught during the Lake Ohrid multimesh gillnets sampling campaign for this project.

4.2. Data analysis and management

Captured fish were identified to species level, counted and weighed in grams. A comprehensive fact sheet of each sampling location has been filled following the previous agreements during the workshops CSBL Project with counting measuring weighting all the fishes per each net. The excel files were later on elaborated following agreement from the reporter leader (in case of Ohrid HBI Ohrid) and in line with outline and guideline provided by the CSBL.

CPUE expressed in biomass of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m2(BPUE) and individuals of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m^2(NPUE).

Figure 16. Fish catch processing

A literature review considered past studies with analytical approaches to the assessment of freshwater fish communities, particularly those relevant to the requirements of CSBL and the WFD.

Species evenness and richness between sub basins were obtained from data and also species length frequency for all species caught.

All processed and analysed data are archived as data sets that can be used for future data base development.

4.3. Results

To adopt the standard EN 14757 from one side and to fulfil the same for large water body as Lake Ohrid is, random sampling with MMGN was performed on seven different (by habitats, wind exposure, water currents, ecological conditions, addition nutrient load and exposure to harmful substances) sub-basins. In total 135 nets were set at both sampling parts of the Lake, 64 in Macedonia and 72 in Albania.

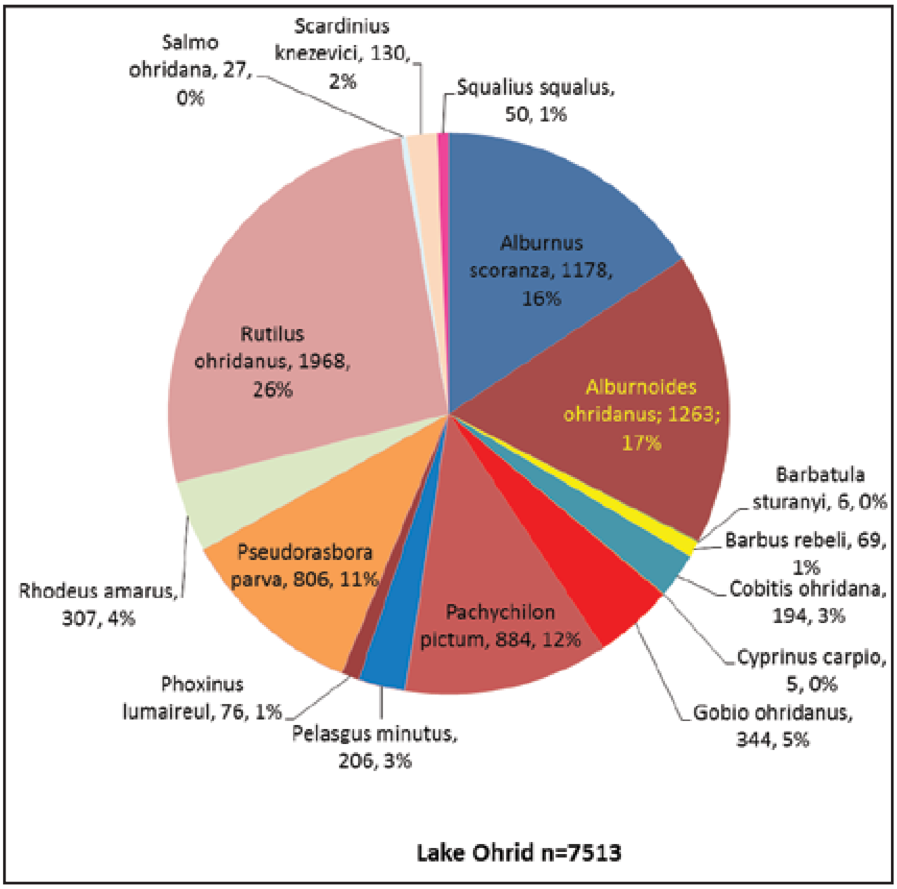
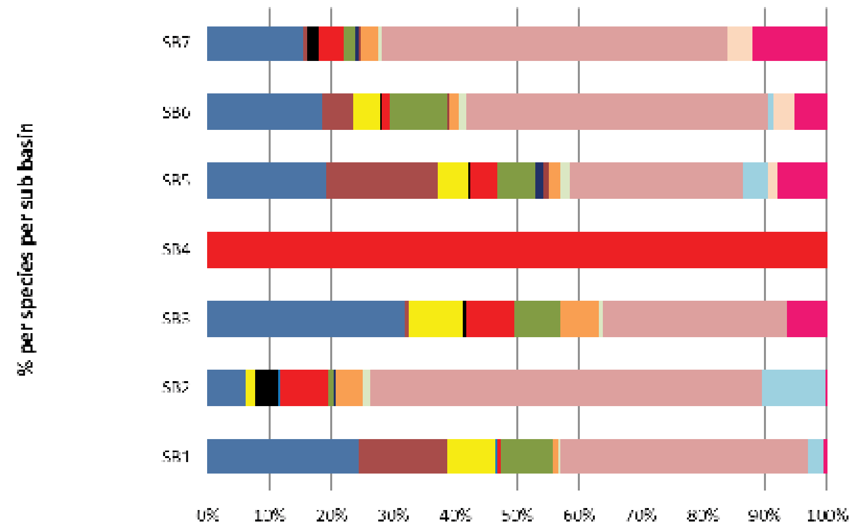


Figure 17. Relative and absolute fish species composition represented in the total catch of Lake Ohrid (SB1-SB7) during the sampling campaign in October-November 2013
(Note: Data label of 0% are representing species whose total number was less than 4 individuals)

This figure in fact represents the actual (instantaneous – ten days sampling campaign) fish assemblage in October and November 2013. In total, 15 fish species were recorded in the catch. These results which are showing dominance of the native species with more than 84% of the total catch can serve mainly about determining the ecological status of certain habitats and also knowledge of the alien species ecology in relation to manage or design certain strategies for their reduction. From other side, regarding fishery issues (catch quotas, minimum catchable size, types of gears or even simple restricted fish stock assessment) the obtained data are not usable.

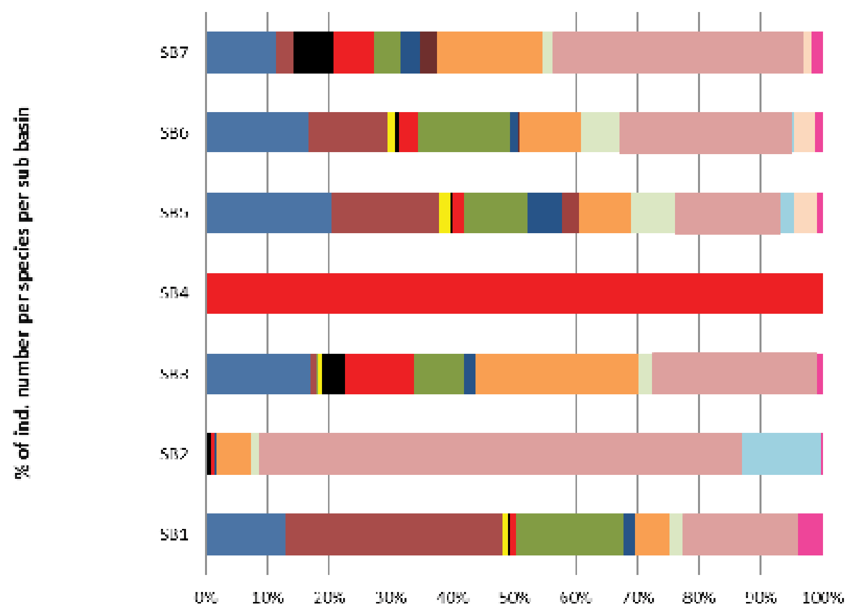


	SB1	SB2	SB3	SB4	SB5	SB6	SB7
Alburnus scoranza	122.54	5.76	36.33		17.58	17.64	10.31
Alburnoides ohridanus	70.81		0.45		16.98	4.67	0.40
Barbatula sturanyi	0.21	0.12	0.03				
Barbus rebeli	39.15	1.54	9.89		4.56	4.26	
Cobitis ohridana	0.38	3.41	0.86		0.14	0.25	1.20
Cyprinus carpio	0.56	0.36					
Gobio ohridanus	4.17	7.25	8.72	1.08	4.10	1.35	2.62
Pachychilon pictum	41.59	1.02	8.14		5.83	8.70	1.23
Pelasgus minutus	0.79	0.13	0.20		1.02	0.19	0.46
Phoxinus lumaireul					0.95	0.03	0.37
Pseudorasbora parva	4.03	4.40	7.02		1.68	1.71	1.88
Rhodeus amarus	1.61	0.91	0.04		1.44	1.20	0.31
Rutilus ohridanus	200.97	60.44	33.90		25.69	46.35	37.36
Salmo ohridanus	11.48	9.66			4.01	0.85	
Scardinius knezevici					1.20	3.43	2.73
Squalius squalus	3.72	0.26	7.27		7.51	4.95	8.14

g/m2

Figure 18. CPUE expressed in biomass (g/m²) in percentage per species of total catch per sub-basin (BPUE) of Lake Ohrid during the sampling campaign in October-November 2013

Taking Lake Ohrid as one water body, regarding the biomass of fish species, three native (bleak roach and spirlin) and two alien species (pumpkinseed and bitterling) are predominant. On the other side, regarding the number of individuals per square meter of net (NPUE) at SB4 only one species Ohrid gudgeon was caught so it is present with 100 %.



	SB1	SB2	SB3	SB4	SB5	SB6	SB7
Alburnus scoranza	2.22	0.10	0.89		1.87	1.55	0.72
Alburnoides ohridanus	6.04		0.05		1.61	1.21	0.18
Barbatula sturanyi	0.02	0.01	0.01				
Barbus rebeli	0.17	0.01	0.04		0.15	0.12	
Cobitis ohridana	0.05	0.47	0.19		0.03	0.05	0.41
Cyprinus carpio	0.01	0.02					
Gobio ohridanus	0.15	0.51	0.60	0.05	0.18	0.40	0.41
Pachychilon pictum	3.02	0.07	0.41		0.96	1.40	0.29
Pelasgus minutus	0.30	0.03	0.09		0.49	0.12	0.19
Phoxinus lumaireul					0.25	0.08	0.18
Pseudorasbora parva	0.97	4.40	1.48		0.79	0.92	1.08
Rhodeus amarus	0.40	0.91	0.13		0.64	0.58	0.11
Rutilus ohridanus	3.16	60.44	1.39		1.58	2.65	2.57
Salmo ohridanus	0.01	9.66			0.20	0.02	
Scardinius knezevici					0.34	0.33	0.09
Squalius squalus	0.67	0.26	0.04		0.05	0.11	0.10

ind./m²

Figure 19. CPUE expressed in number of individuals/m² in percentage per species of total catch per sub-basin (NPUE) of Lake Ohrid during the sampling campaign in October-November 2013

4.3.1. Multimesh gillnetting

4.3.1.1. SB1 – Veli Dab

The sampling at SB1 took time within two campaigns. This sub basin from other hand is representing spawning ground in spring and summer for a lot of fishes from the Cyprinidae family, and in winter for the salmonids. Hence, choosing the period of second half of October, gives possibility to realize the CEN standard as much as appropriate, in terms of non-aggregated fish populations. This SB, along the shore and towards the deep, differs very much in microhabitat's conditions. From other hand, this issue was also as a condition for delineation of this sub basin. Interesting for this sub basin is the presence of numerous sublacustrine springs.

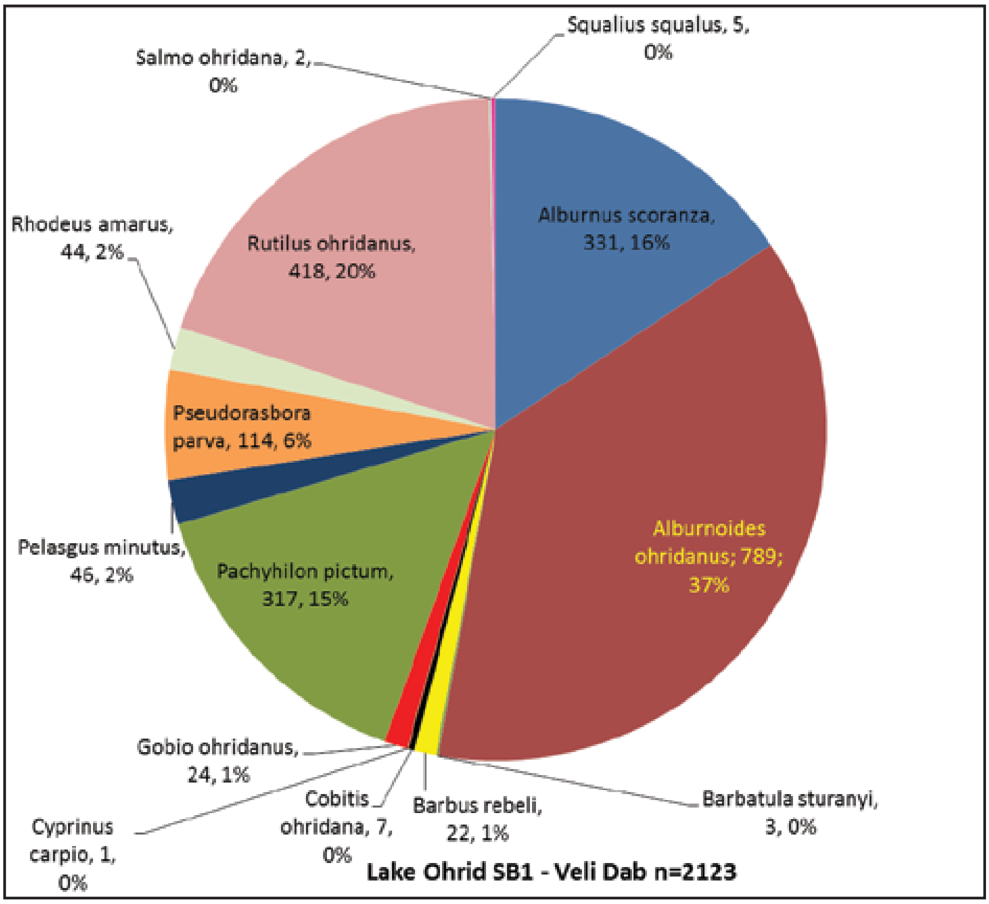
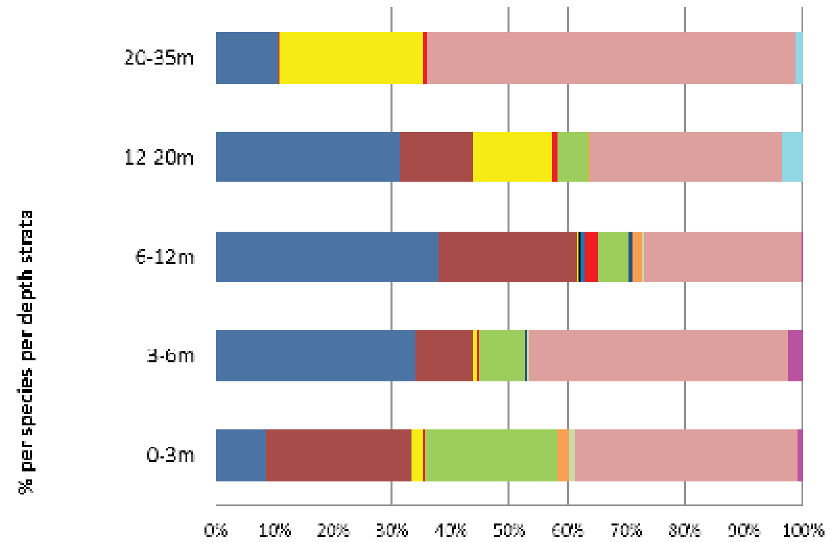


Figure 20. Relative fish species composition in total catch at SB1 in the sampling campaign October-November 2013 (Note: Data label of 0% are representing species whose total number was less than 7 individuals)

From the sampling at this sub basin, the results showed species dominance of mostly noncommercial Ohrid spirlin and minnow – pachychilon or low valued commercial species roach and bleak. Although, the fishing was performed even at rather deep strata, the catch of the commercially valued species was negligible - 2 individuals of the endemic Lake Ohrid belvica and 1 carp (Figure 20). At this sub basin totally 14 fish species were caught.



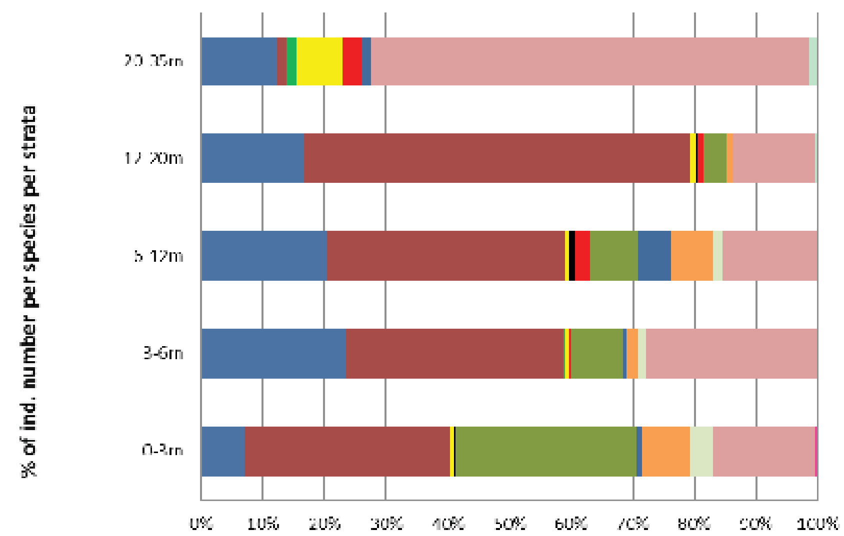
	0-3m	3-6m	6-12m	12-20m	20-35m
Alburnus scoranza	8.38	34.83	38.07	31.51	9.75
Alburnoides ohridanus	25.06	9.86	23.35	12.27	0.27
Barbatula sturanyi	0.00	0.04	0.06	0.00	0.11
Barbus rebeli	1.93	0.60	0.54	13.49	22.58
Cobitis ohridana	0.03	0.00	0.26	0.10	0.00
Cyprinus carpio	0.00	0.00	0.56	0.00	0.00
Gobio ohridanus	0.12	0.42	2.23	0.80	0.60
Pachychilon pictum	22.67	8.18	5.33	5.42	0.00
Pelasgus minutus	0.15	0.06	0.53	0.00	0.05
Pseudorasbora parva	1.96	0.24	1.67	0.16	0.00
Rhodeus amarus	0.91	0.31	0.40	0.00	0.00
Rutilus ohridanus	37.83	45.06	26.66	32.87	58.55
Salmo ohridana	0.00	0.00	0.00	3.38	1.00
Squalius squalus	0.96	2.42	0.34	0.00	0.00

g/m²

Figure 21. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 1 locality

Regarding CPUE for the same sub basin, the dominance in biomass is different due to species sizes and numbers caught here.

Therefore, the roach was dominant in almost the whole range of the fishing strata (0-50 m) with interesting biomass distribution progressively from the shore to the deep and almost opposite for the bleak from the deep to the shore. The presence of the endemic Ohrid minnow (Pachychillon) and the Ohrid spirling are almost following the presence in biomass like the rest of the dominant ones in this sub basin.



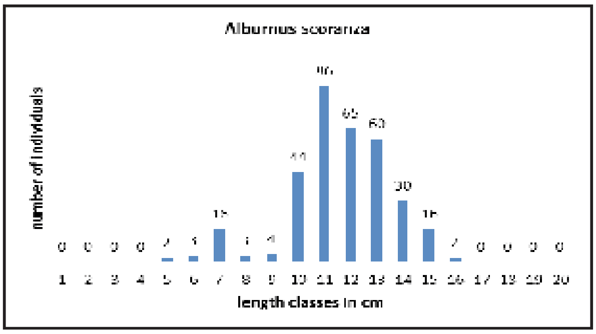
	0-3m	3-6m	6-12m	12-20m	20-35m
Alburnus scoranza	0.60	0.54	0.76	0.76	0.06
Alburnoides ohridanus	2.82	0.81	1.42	0.98	0.01
Barbatula sturanyi	0.00	0.01	0.01	0.00	0.01
Barbus rebeli	0.08	0.07	0.03	0.01	0.04
Cobitis ohridana	0.01	0.00	0.03	0.01	0.00
Cyprinus carpio	0.00	0.00	0.01	0.00	0.00
Gobio ohridanus	0.01	0.01	0.09	0.01	0.01
Pachychilon pictum	2.49	0.19	0.78	0.06	0.00
Pelasgus minutus	0.08	0.02	0.19	0.00	0.01
Pseudorasbora parva	0.67	0.04	0.25	0.01	0.00
Rhodeus amarus	0.31	0.03	0.06	0.00	0.00
Rutilus ohridanus	1.41	0.64	0.57	0.21	0.34
Salmo ohridana	0.00	0.00	0.00	0.01	0.01
Squalius squalus	0.03	0.01	0.01	0.00	0.00

ind./m²

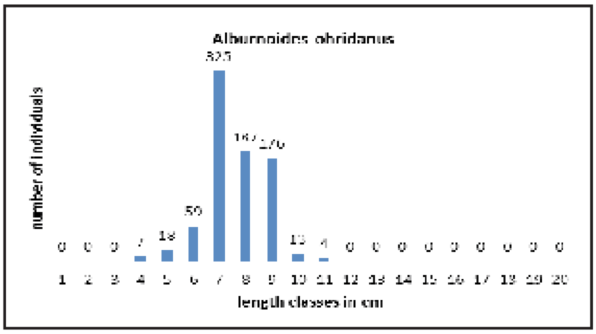
Figure 22. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 1 locality

In terms of the fish distribution per depth expressed in number of individuals, the roach shows biggest presence in the depth strata of 20 to 35 m. The bleak population distribution is almost the same as the biomass distribution per depth strata. The Ohrid spirlin occupies almost all the presented depth strata from 0-20 m depth.

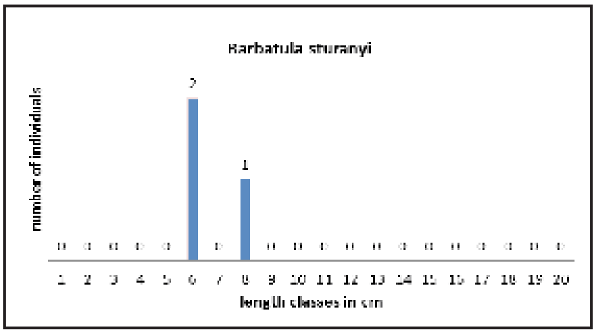
The length frequency distributions of the fish species caught during the survey at the SB 1 are presented in the following figure (23; a-n).



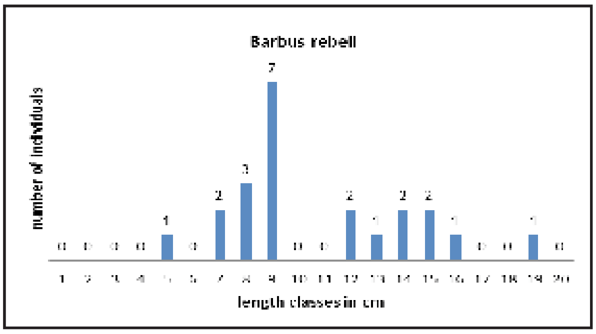
a)



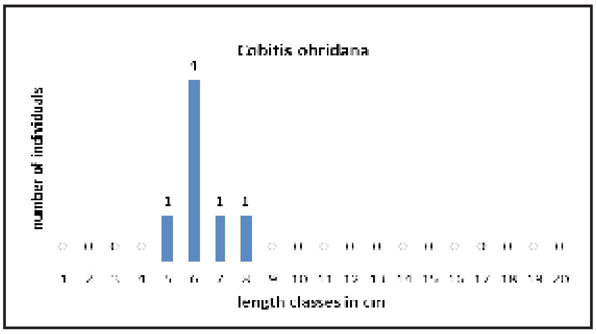
b)


Barbatula sturanyi
number of individuals
length classes in cm

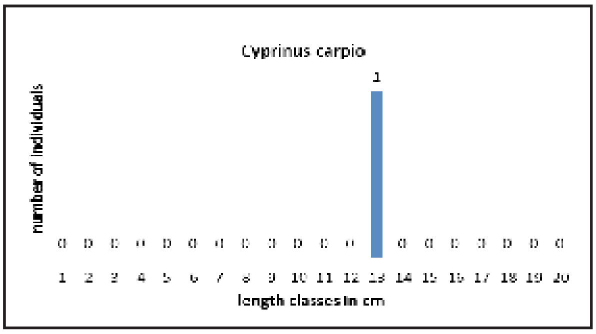
c)


Barbus rebeli
number of individuals
length classes in cm

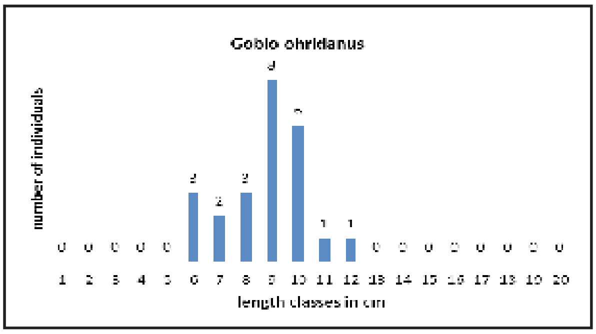
d)


Cobitis ohridana
number of individuals
length classes in cm

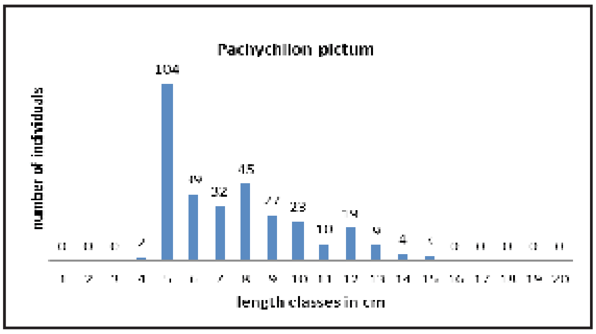
e)


Cyprinus carpio
number of individuals
length classes in cm

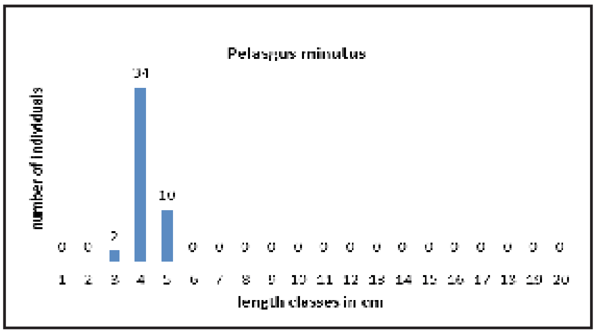
f)


Gobio ohridanus
number of individuals
length classes in cm

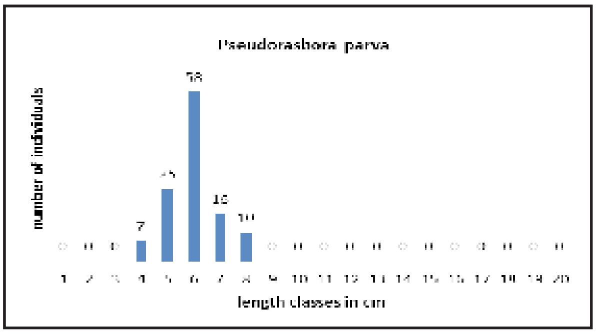
g)


Pachychilon pictum
number of individuals
length classes in cm

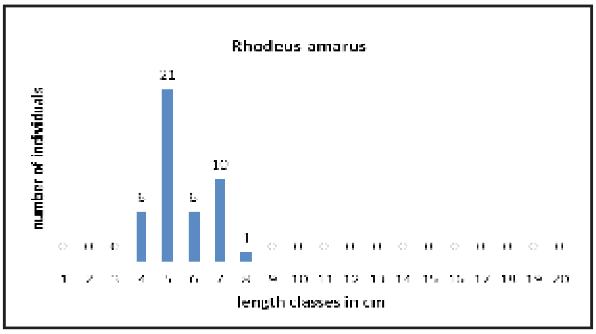
h)


Pelasgus minutus
number of individuals
length classes in cm

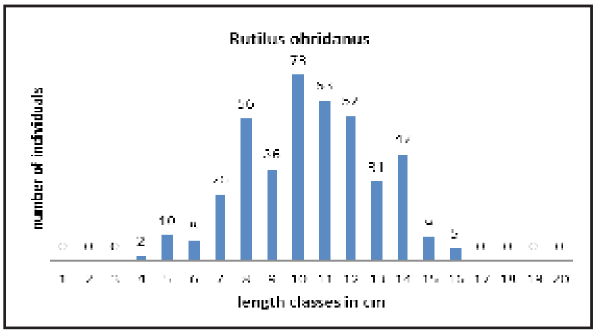
i)


Pseudorasbora parva
number of individuals
length classes in cm

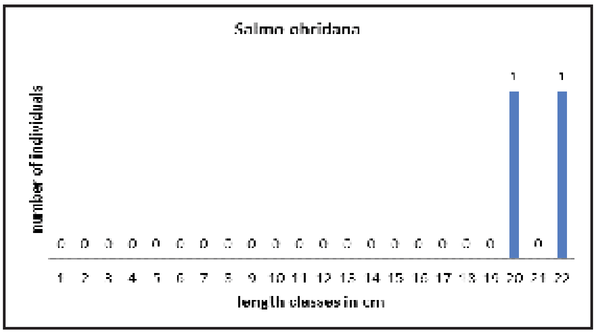
j)



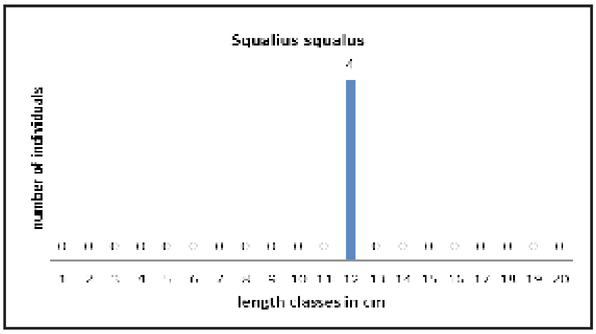
k)



l)



m)



n)

Figure 23. Length classes frequency per species for the SB1 Veli Dab (a-n)

From the above graphs, it is worth to mention that all the native fish minnows are represented with their whole age structure.

The structure of the bleak and the roach population gives normal size and age distribution for this period of the year. Thus, the roach is represented with 14 length classes and the bleak with 12. Significant to mention is the number of the 10 length classes of the Ohrid barbell. But regarding the older knowledge about its presence at this sub basin from the fishery statistics of the fishing company, it appears that the barbell population is improving at this region.

Also the significant presence of the alien species stone moroko and bitterling with their relative abundance for this sub basin gives new results – previously not recorded at this region..

The most important is that the salmonid species were very insignificantly represented and only Lake Ohrid belvica with 2 specimens, which may be result of relatively unfavorable water temperature, higher than 15 ^{0}C or of the type of the fishing gear used..

But, nevertheless this exercise, with CEN standard and such MMG, used for the first time at this sub basin, gives an interesting and useful data about some species distribution, mainly concerning the alien ones. Thus, further improving of the whole CEN scheme as in sampling as well in gears adjusted for Lake Ohrid condition can produce a substrate for developing new Fish Monitoring standards for big and deep lakes.

4.3.1.2. SB2 – Andon Dukov

The SB2 at the region named according the auto-camping site "Andon Dukov", represents in terms of habitat characterization, sandy bottom, with a reed belt of *Phragmites australis* from the shore till depth of 1,2 m, while patches of *Chara sp, Myrophylum sp. and Potamogeton sp.* are scattered till a depth of 8 m depth. In the sandy bottom representatives from the mussel *Unio sp.* are significantly present. The bottom slope at this SB2, unlike SB1, is lower with sublacustrine terrace starting 600 m from the shoreline. As in this fine sand/muddy bottom quite a lot of macrozoobenthos species are present , accordingly to the fish species diet, the graph bellow represents an ecosystem characteristics of SB2 including the fish fauna as an integrative part.

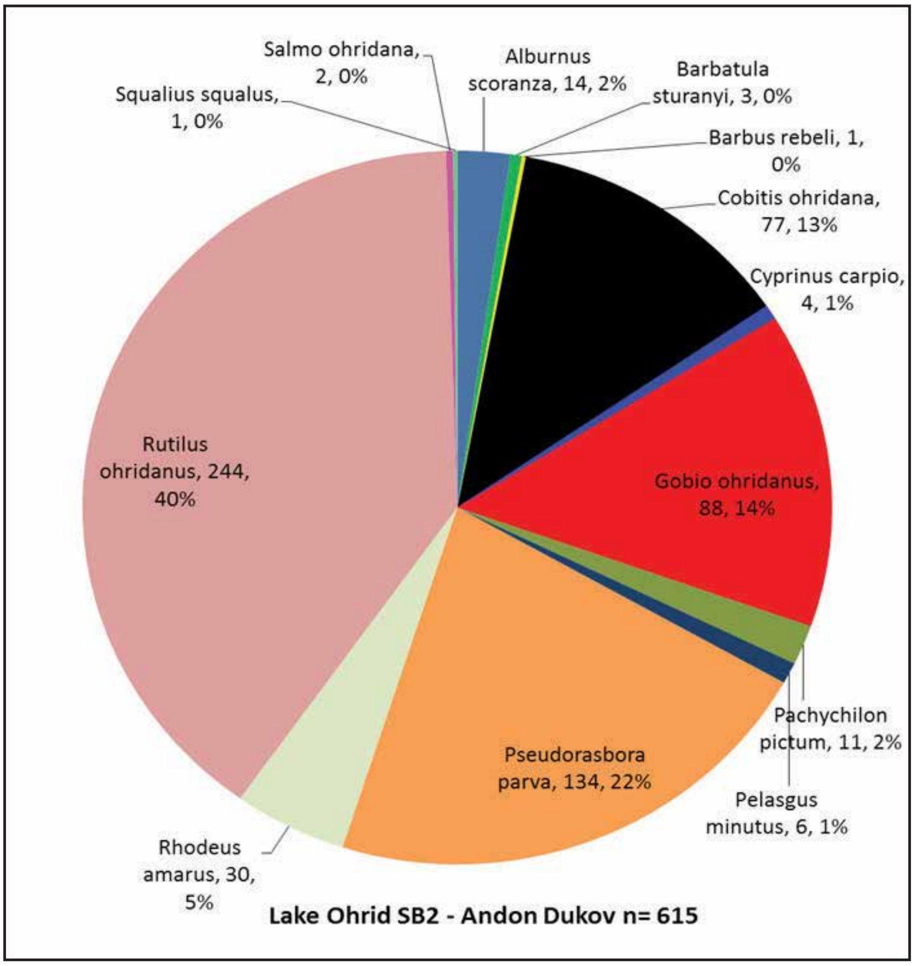
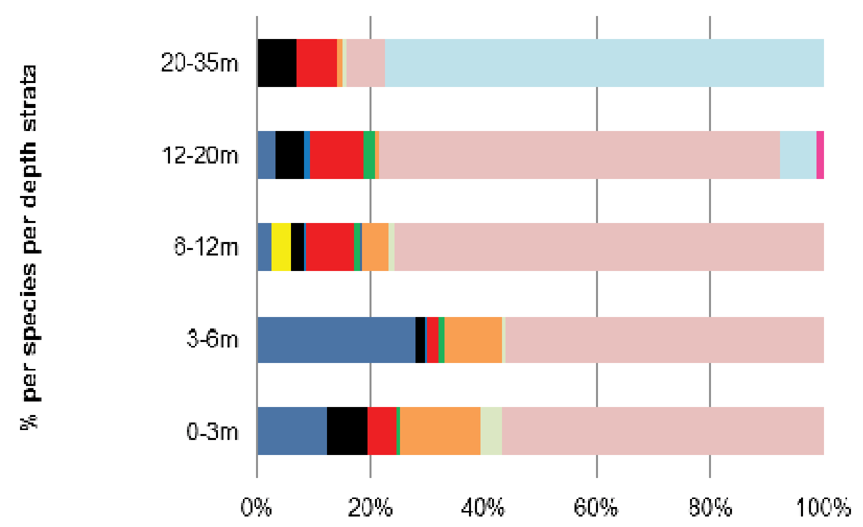


Figure 24. Relative fish species composition in total catch at SB2 in the sampling campaign October-November 2013 (Note: Data label of 0% are representing species whose total number was less than 3 individuals)

At this sub basin, is obvious the dominance of the roach and other zoobenthofagous species, where amongst the native species, significant presence of the aliens stone moroko and bitterling is evident. At this sub basin totally 13 fish species were caught.

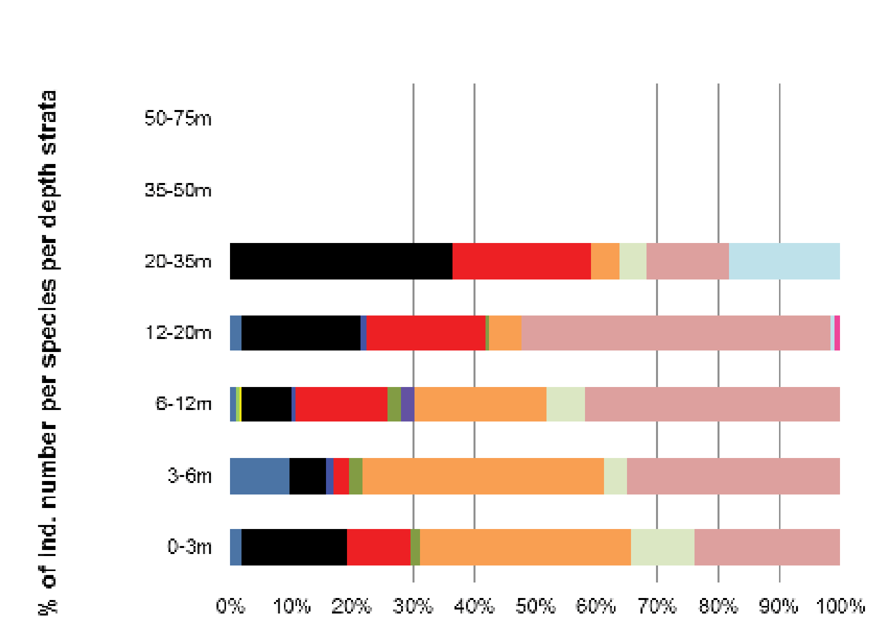


	0-3m	3-6m	6-12m	12-20m	20-35m
Alburnus scoranza	0,60	3,33	1,17	0,66	0,00
Barbatula sturanyi	0,00	0,00	0,12	0,00	0,00
Barbus rebeli	0,00	0,00	1,54	0,00	0,00
Cobitis ohridana	0,34	0,23	1,07	1,01	0,76
Cyprinus carpio	0,00	0,04	0,13	0,18	0,00
Gobio ohridanus	0,24	0,21	4,10	1,91	0,77
Pachychilon pictum	0,04	0,14	0,46	0,38	0,00
Pelasgus minutus	0,00	0,00	0,13	0,00	0,00
Pseudorasbora parva	0,68	1,21	2,26	0,16	0,09
Rhodeus amarus	0,18	0,07	0,60	0,00	0,06
Rutilus ohridanus	2,75	6,72	35,94	14,26	0,76
Salmo ohridana	0,00	0,00	0,00	1,27	8,38
Squalius squalus	0,00	0,00	0,00	0,26	0,00

g/m²

Figure 25. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 2 locality

Regarding CPUE for the same sub basin, the dominance in biomass is also expressed on roach, whilst Ohrid belvica (with 5 individuals) dominates in the strata from 20-35 m.



	0-3m	3-6m	6-12m	12-20m	20-35m	35-50m	50-75m
Alburnus scoranza	0,01	0,06	0,02	0,01	0,00	0	0
Barbatula sturanyi	0,00	0,00	0,01	0,00	0,00	0	0
Barbus rebeli	0,00	0,00	0,01	0,00	0,00	0	0
Cobitis ohridana	0,11	0,04	0,14	0,12	0,06	0	0
Cyprinus carpio	0,00	0,01	0,01	0,01	0,00	0	0
Gobio ohridanus	0,07	0,01	0,27	0,12	0,04	0	0
Pachychilon pictum	0,01	0,01	0,04	0,01	0,00	0	0
Pelasgus minutus	0,00	0,00	0,03	0,00	0,00	0	0
Pseudorasbora parva	0,22	0,24	0,39	0,03	0,01	0	0
Rhodeus amarus	0,07	0,02	0,11	0,00	0,01	0	0
Rutilus ohridanus	0,16	0,21	0,74	0,32	0,02	0	0
Salmo ohridana	0,00	0,00	0,00	0,01	0,03	0	0
Squalius squalus	0,00	0,00	0,00	0,01	0,00	0	0

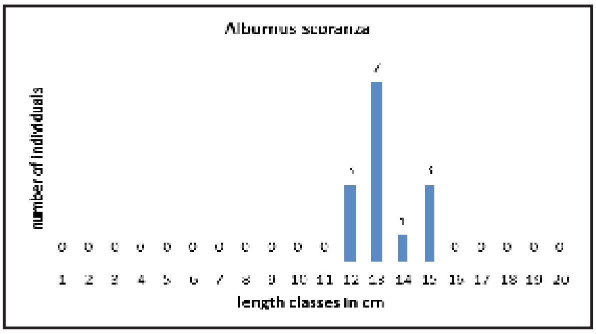
ind./m²

Figure 26. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 2 locality

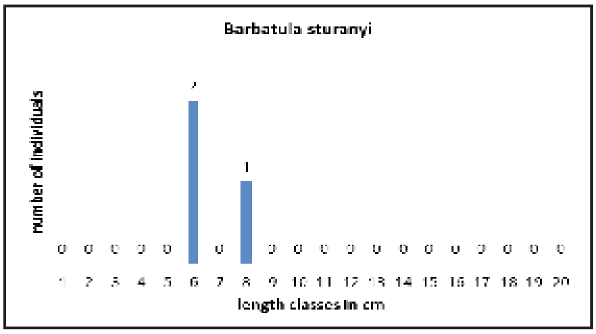
Unlike the biomass distribution per depth strata for this sub basin 2, the presence in number of fish species per m² is different. Here apart from the dominancy of the roach, the spine and stone loaches from the natives and stone moroko are relatively high represented. Having in mind the ecological conditions at this sub basin (the bottom substrate and macrophyte vegetation) such kind of distribution could be expected as also previously described by Spirkovski et all. 2002.

The two depth strata 20-35 and 35-50m in this sub basin remain without any catch although nets were set in them.

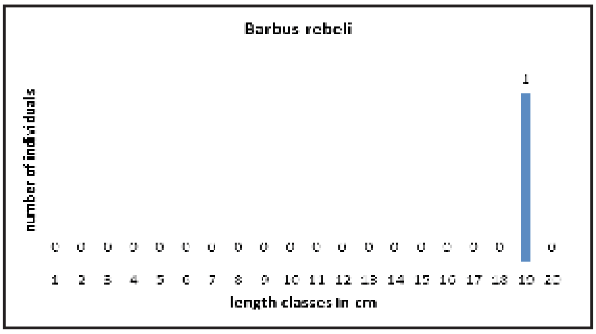
The length frequency distributions of the fish species caught during the survey at the SB 2 are presented in the following figure (27; a-m).


Alburnus scoranza
number of individuals
length classes in cm

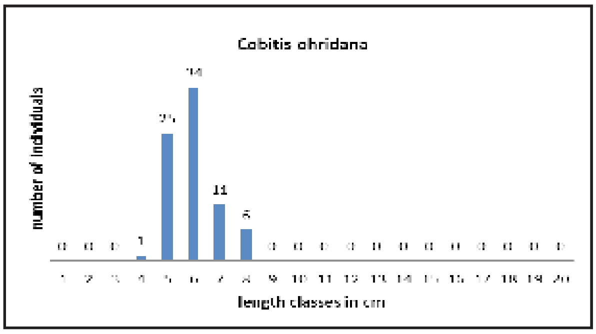
a)


Barbatula sturanyi
number of individuals
length classes in cm

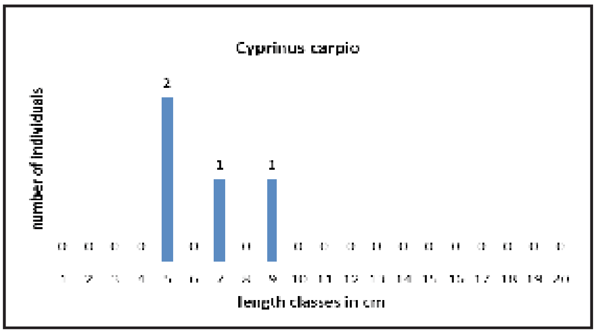
b)


Barbus rebeli
number of individuals
length classes in cm

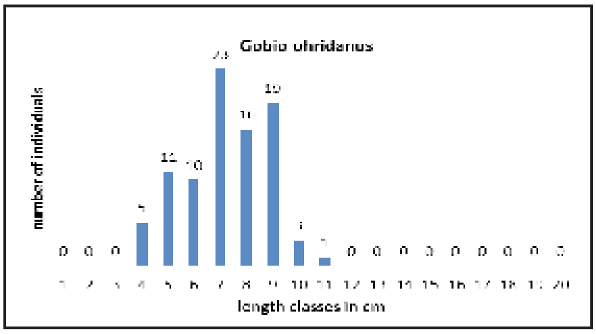
c)


Cobitis ohridana
number of individuals
length classes in cm

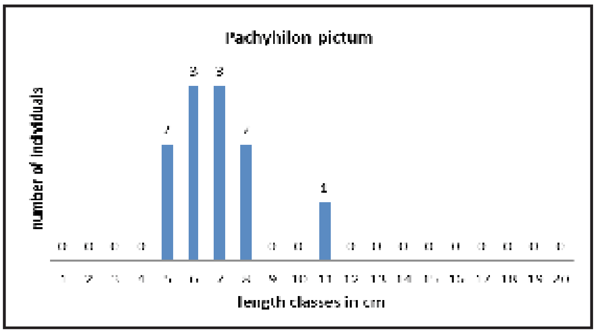
d)


Cyprinus carpio
number of individuals
length classes in cm

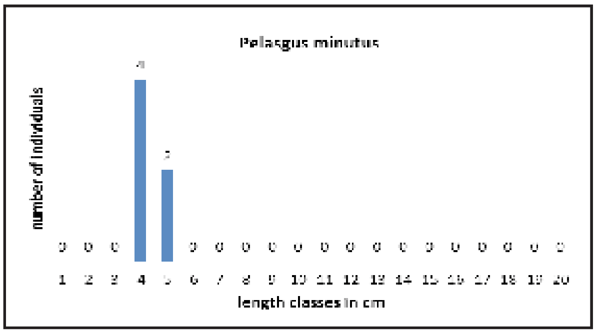
e)


Gobio ohridanus
number of individuals
length classes in cm

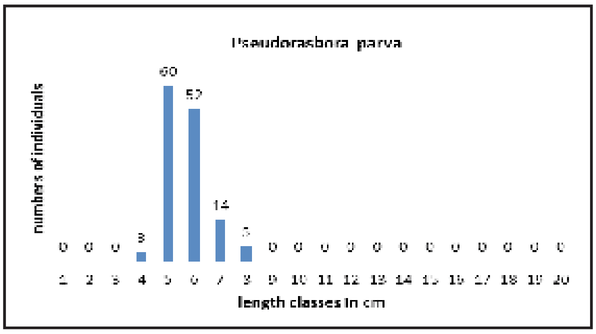
f)


Pachychilon pictum
number of individuals
length classes in cm

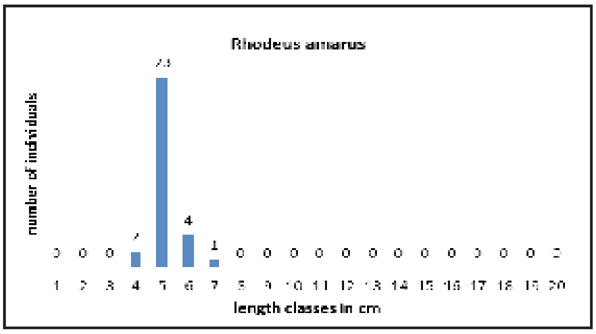
g)


Pelasgus minutus
number of individuals
length classes in cm

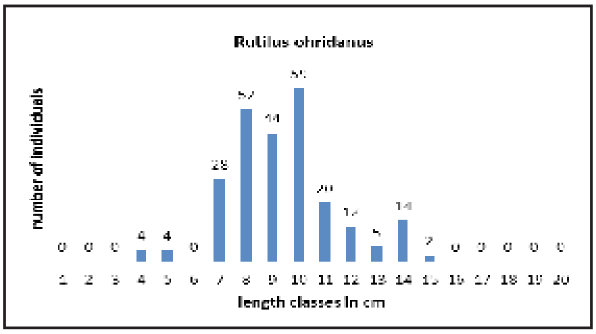
h)



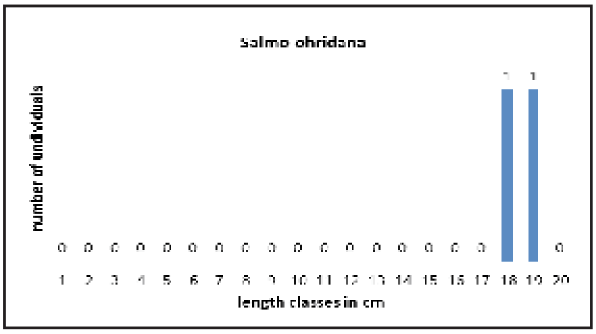
i)



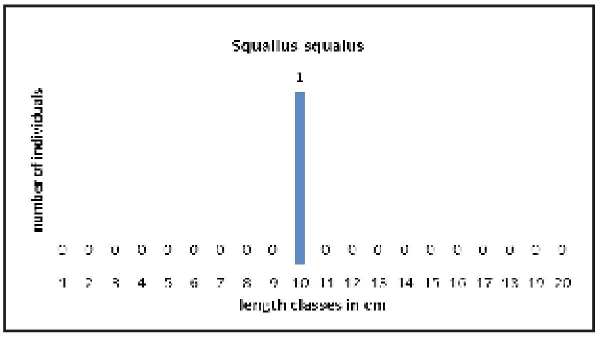
j)



k)



l)



m)

Figure 27. Length classes frequency per species for the SB2 Andon Dukov (a-m)

From the above graphs, it can be seen that only few commercial species were present in the whole catch meaning on roach (mainly) and belvica with barbell barely.

Same like for the previous SB1 using CEN standard and such MMG, for the first time, gives an interesting and useful data about some species distribution. From the length classes of the native fishes dominates the roach with 11 sizes, and followed by the Ohrid gudgeon with eight classes.

4.3.1.3. SB3 – Radozda

The SB3 around the village of Radozda on the West shore of the Macedonian part of the lake, from its habitat characteristics represents a kind of mixture of the two previous SB1 and SB2. That was also main attribute for delineation of this water sub basin for the purpose of this project, from the fishery point.

The sampling at this SB took part at the beginning of November 2013.

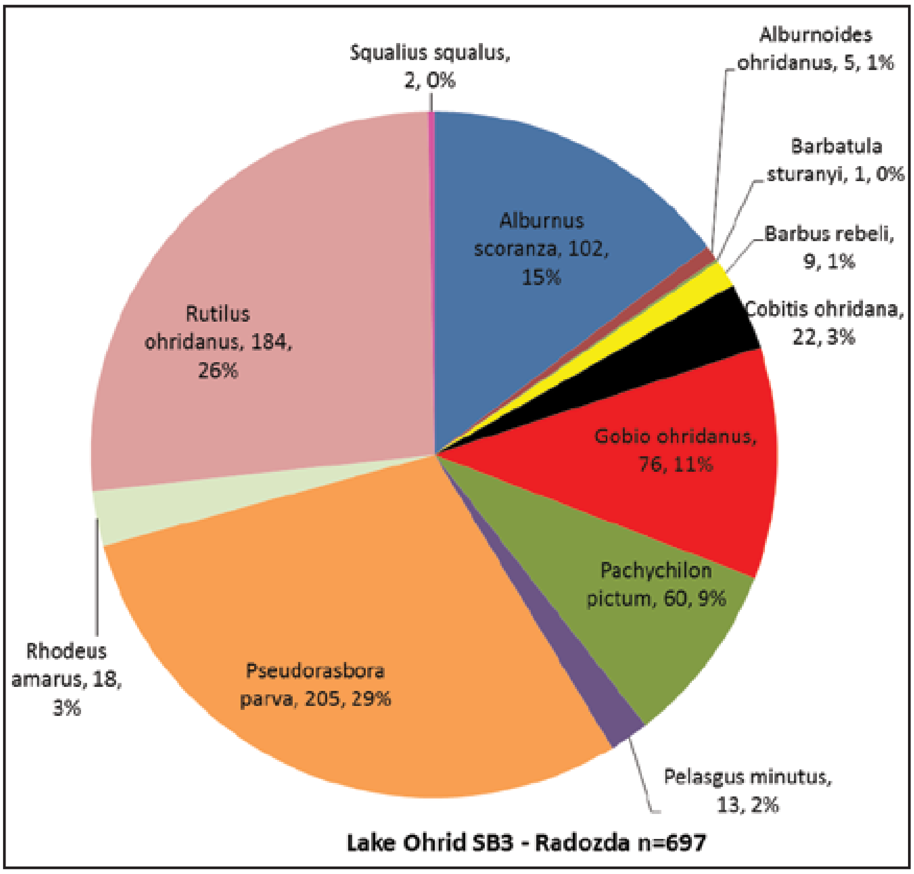
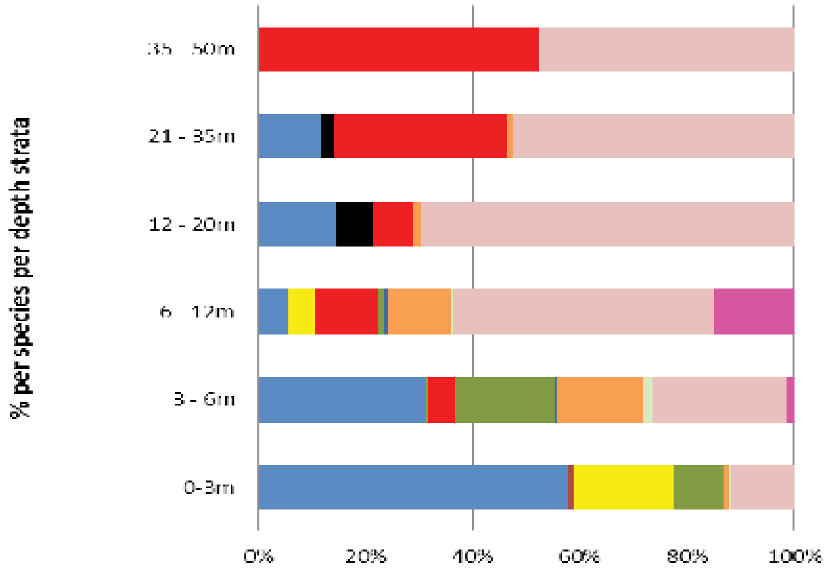


Figure 28. Relative fish species composition in total catch at SB3 in the sampling campaign November 2013 (Note: Data label of 0% are representing species whose total number was less than 2 individuals)

From the sampling at this sub basin, the results showed dominance of commercially non valued species (minnows and alien, stone moroko) and whilst the low commercially valued species like bleak and roach represented together with 41% , the more attractive fish were represented only with 6 barbell specimens. All together 12 fish species were caught.



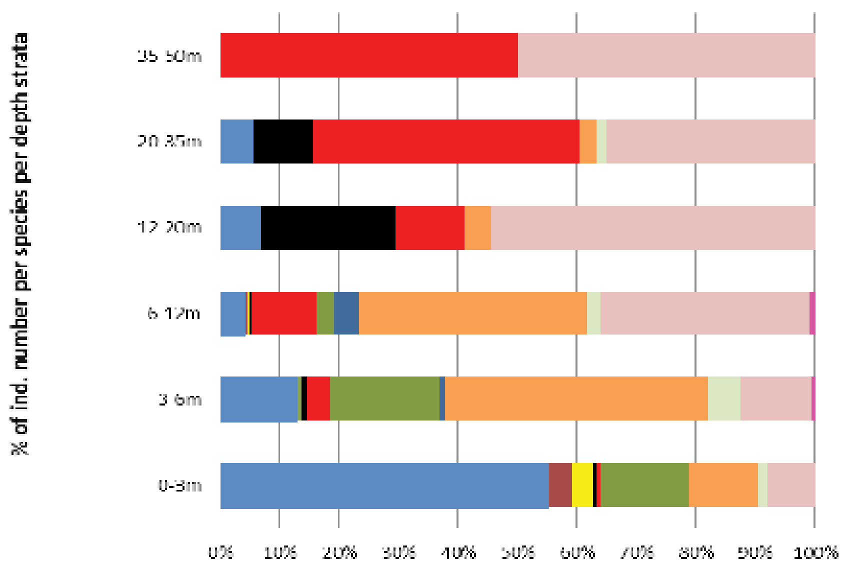
	0-3m	3 - 6m	6 - 12m	12 - 20m	21 - 35m	35 - 50m
Alburnus scoranza	26.39	6.24	1.42	0.87	1.40	0.00
Alburnoides ohridanus	0.43	0.00	0.02	0.00	0.00	0.00
Barbatula sturanyi	0.00	0.03	0.00	0.00	0.00	0.00
Barbus rebeli	8.52	0.00	1.36	0.00	0.00	0.00
Cobitis ohridana	0.05	0.06	0.05	0.42	0.29	0.00
Gobio ohridanus	0.06	1.02	3.04	0.44	3.87	0.28
Pachyhilon pictum	4.19	3.71	0.24	0.00	0.00	0.00
Pelasgus minutus	0.00	0.04	0.17	0.00	0.00	0.00
Pseudorasbora parva	0.48	3.19	3.16	0.11	0.09	0.00
Rhodeus amarus	0.13	0.35	0.12	0.00	0.04	0.00
Rutilus ohridanus	5.40	4.98	12.77	4.22	6.27	0.25
Squalius squalus	0.00	0.30	3.98	0.00	0.00	0.00

g/m²

Figure 29. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 3 locality

Regarding CPUE for this sub basin, the dominance in biomass is almost equal to the dominance in number represented in the catch.

Thus, the roach, also here, is the leading one in almost all presented depth strata. The bleak dominates significantly in the shallower strata 0-6 m. The Ohrid gudgeon is significantly represented in the last two deepest strata.



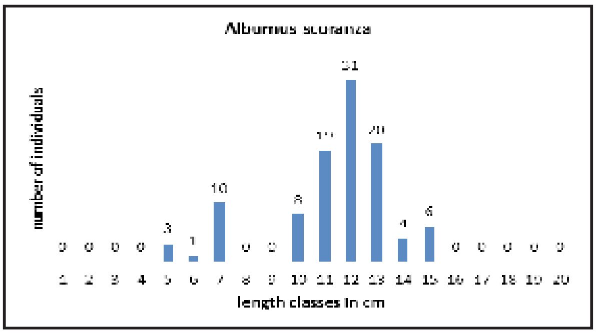
	0-3m	3-6m	6-12m	12-20m	20-35m	35-50m
Alburnus scoranza	0.6	0.14	0.08	0.03	0.03	0
Alburnoides ohridanus	0.04	0.00	0.01	0.00	0.00	0.00
Barbatula sturanyi	0.00	0.01	0.00	0.00	0.00	0.00
Barbus rebeli	0.04	0.00	0.01	0.00	0.00	0.00
Cobitis ohridana	0.01	0.01	0.01	0.11	0.05	0.00
Gobio ohridanus	0.01	0.04	0.21	0.06	0.24	0.04
Pachychilon pictum	0.16	0.20	0.05	0.00	0.00	0.00
Pelasgus minutus	0.00	0.01	0.08	0.00	0.00	0.00
Pseudorasbora parva	0.13	0.49	0.73	0.02	0.01	0.00
Rhodeus amarus	0.01	0.06	0.04	0.00	0.01	0.00
Rutilus ohridanus	0.09	0.13	0.67	0.27	0.19	0.04
Squalius squalius	0.00	0.01	0.01	0.00	0.00	0.00

ind./m²

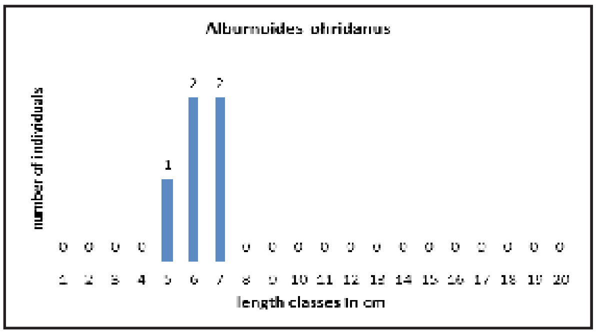
Figure 30. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 3 locality

In terms of presence of individuals number per depth strata, in this case of SB3 the distribution is similar as in the graph for the biomass distribution.

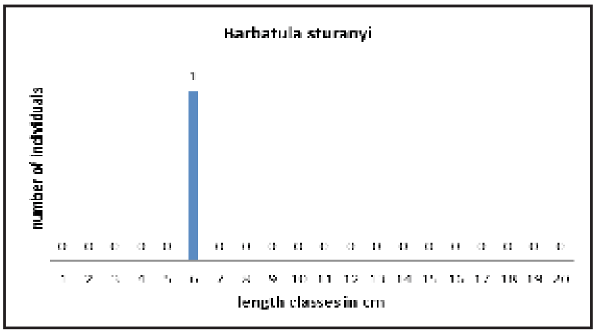
The length frequency distributions of the fish species caught during the survey at the SB 3 are presented in the following figure (31; a-l).



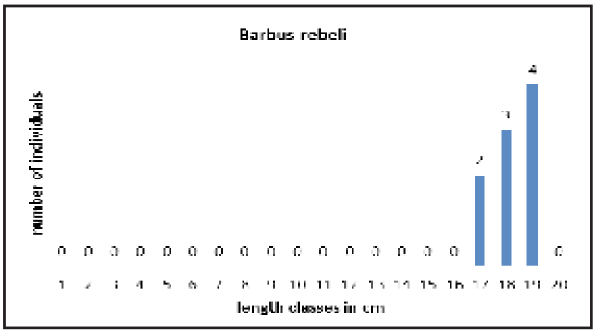
a)



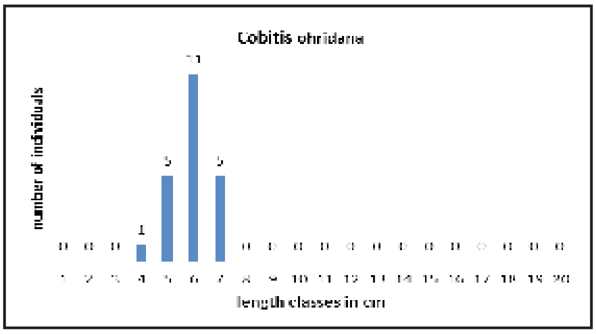
b)


Barbatula sturanyi
number of individuals
length classes in cm

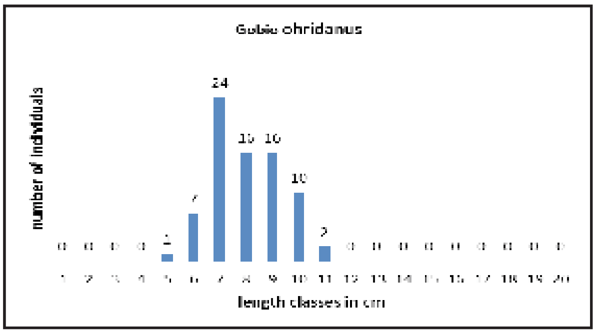
c)


Barbus rebeli
number of individuals
length classes in cm

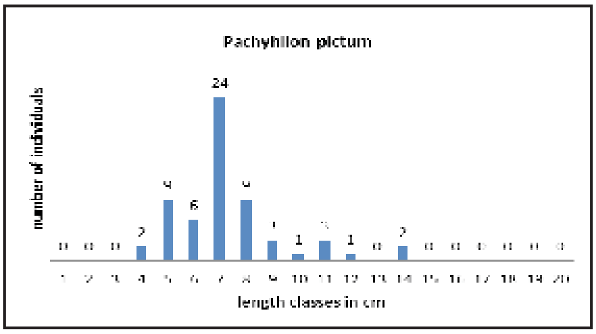
d)


Cobitis ohridana
number of individuals
length classes in cm

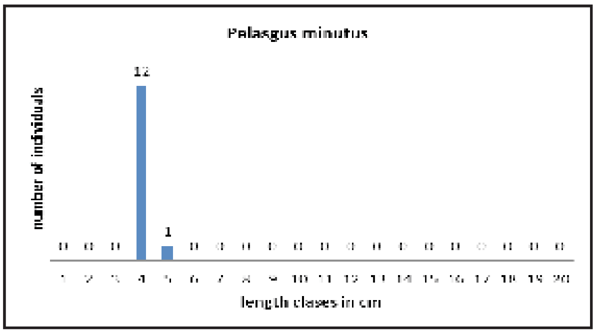
e)


Gobio ohridanus
number of individuals
length classes in cm

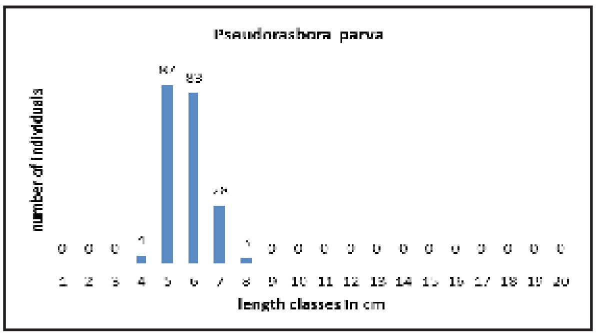
f)


Pachyhilon pictum
number of individuals
length classes in cm

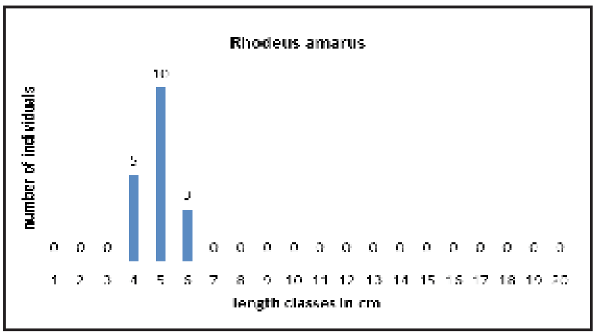
g)


Pelasgus minutus
number of individuals
length classes in cm

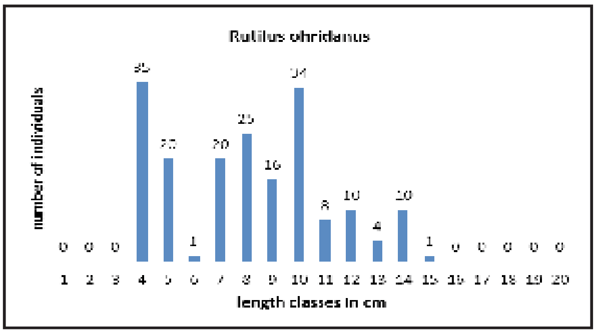
h)


Pseudorasbora parva
number of individuals
length classes in cm

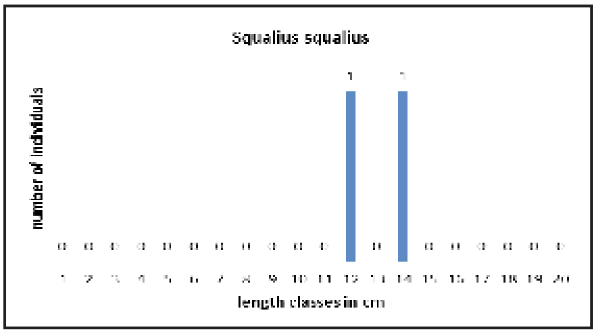
i)


Rhodeus amarus
number of individuals
length classes in cm

j)



k)



l)

Figure 31. Length classes frequency per species for the SB3 Radozda (a-l)

From the above graphs are, more or less showing the presence of same age cohorts among the caught fish species. Thus, in this case also the roach is most dominant with 12 length classes, followed with 10 of the Ohrid minnow (*Pachychilon*) and 9 of bleak length classes.

4.3.1.4. SB4 – Central plate

At the pelagic sub-basin only one species - Ohrid gudgeon with 23 individuals were caught in 10 nets.

The length frequency distribution of this species caught during the survey at the SB 4 is presented in the following figure.

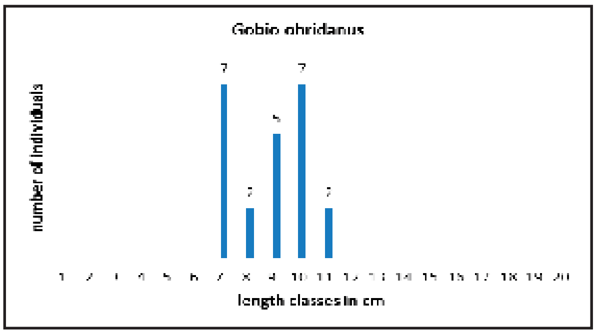


Figure 32. Length classes frequency per species for the SB4 Central plate

The graphs are showing the presence of Ohrid gudgeon in bigger depths, or almost all over the whole bottom strata of Lake Ohrid as described previously by Tocko (1987).

4.3.1.5. SB5 – Lin – Bakalice

The sampling at SB5 took time within three campaigns. This SB, along the shore and towards the deep, differs very much in microhabitat's conditions, and it represent the main spawning ground in winter for the salmonids and in spring and summer for common carp and other species of Cyprinid family.

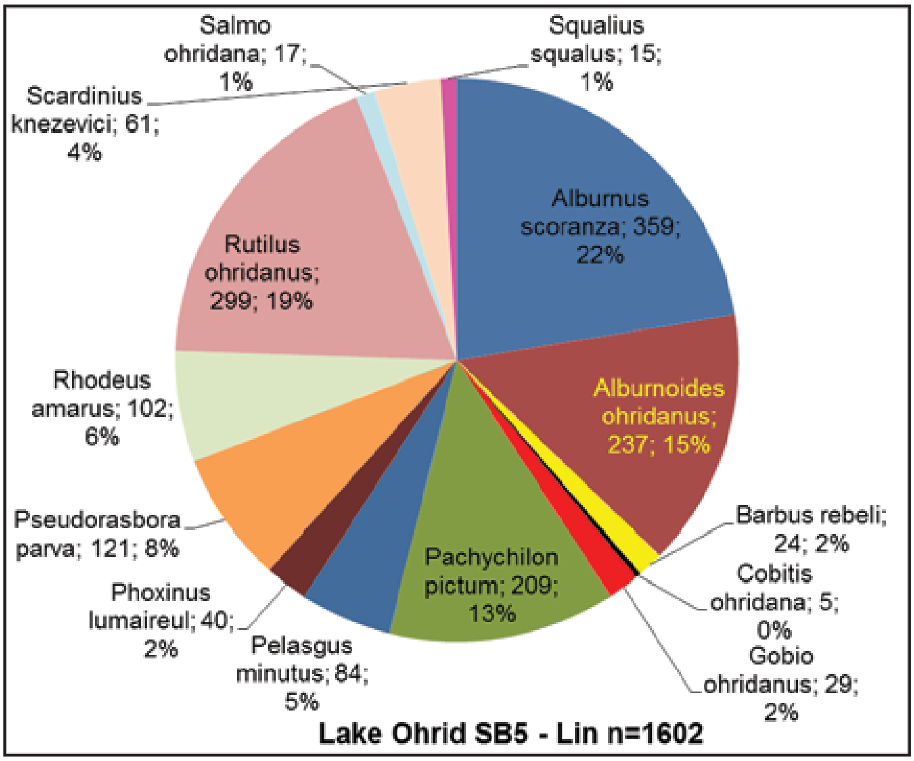
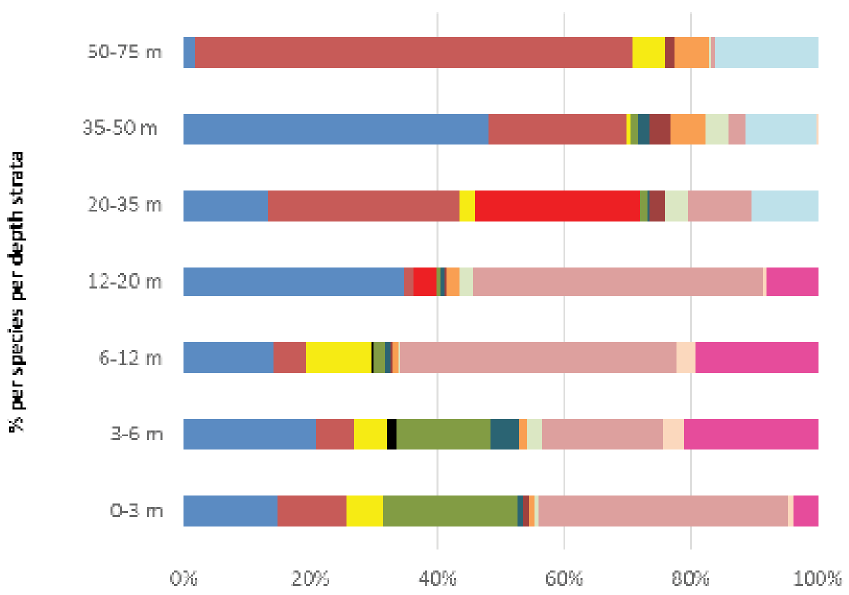


Figure 33. Relative fish species composition in SP 1 (Lin- Bakalice) in the sampling campaign November 2013

From the sampling at sub basin 5, the results showed the dominance of low valued commercial species bleak and roach or noncommercial species Ohrid minnow-Pachychilon and spirlin. Although, the fishing was performed even at rather deep strata, there was no presence of the commercially valued species. At this sub basin totally 13 fish species were caught.

Unlike the previous described sub basins belonging to the Macedonian part, at this sub basin the rudd (*Scardinius knezevici*) appears in the catches.

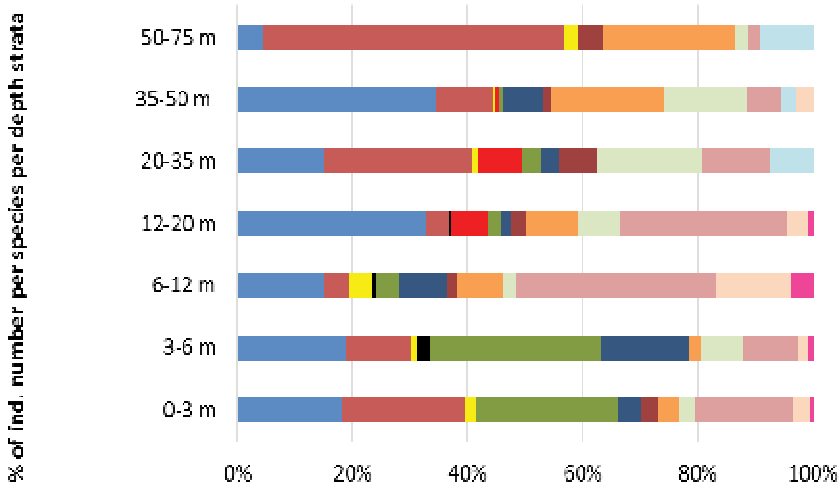


	0-3 m	3-6 m	6-12 m	12-20 m	20-35 m	35-50 m	50-75 m
Alburnus scoranza	2.74	1.55	3.05	4.49	1.85	3.67	0.20
Alburnoides ohridanus	2.05	0.46	1.13	0.20	4.20	1.60	7.25
Barbus rebeli	1.03	0.40	2.18	0.00	0.36	0.03	0.56
Cobitis ohridana	0.00	0.05	0.04	0.01	0.00	0.00	0.00
Gobio ohridanus	0.00	0.00	0.00	0.45	3.64	0.00	0.00
Pachychilon pictum	3.95	1.13	0.44	0.10	0.12	0.10	0.00
Pelasgus minutus	0.20	0.37	0.17	0.06	0.08	0.15	0.00
Phoxinus lumaireul	0.15	0.00	0.04	0.06	0.13	0.75	0.13
Pseudorasbora parva	0.16	0.09	0.17	0.26	0.00	0.41	0.60
Rhodeus amarus	0.11	0.19	0.08	0.26	0.48	0.28	0.08
Rutilus ohridanus	7.26	1.45	9.36	5.93	1.42	0.21	0.06
Salmo ohridana	0.00	0.00	0.00	0.00	1.45	0.85	1.70
Scardinius knezevici	0.15	0.26	0.66	0.07	0.00	0.02	0.00
Squalius squalus	0.78	1.60	4.13	1.06	0.00	0.00	0.00

g/m²

Figure 34. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 5 locality

Regarding this sub basin 5 the distribution of the biomass is with dominance of the roach and bleak; till 20 m depth for the first and till 50m for the second species respectfully. Significant biomass presence is the one from the chub, which differs from the previously described sub basins. Another fact is remarkable for this region that the presence of Ohrid spirlin in such deep strata (50-75m) is recorded.



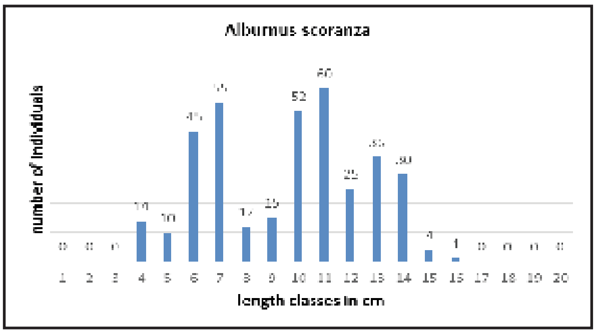
	0-3 m	3-6 m	6-12 m	12-20 m	20-35 m	35-50 m	50-75 m
Alburnus scoranza	0.42	0.17	0.20	0.47	0.16	0.41	0.04
Alburnoides ohridanus	0.50	0.10	0.06	0.06	0.27	0.12	0.51
Barbus rebeli	0.05	0.01	0.05	0.00	0.01	0.01	0.02
Cobitis ohridana	0.00	0.02	0.01	0.00	0.00	0.00	0.00
Gobio ohridanus	0.00	0.00	0.00	0.09	0.08	0.01	0.00
Pachychilon pictum	0.57	0.27	0.05	0.03	0.03	0.01	0.00
Pelasgus minutus	0.10	0.14	0.11	0.03	0.03	0.08	0.00
Phoxinus lumaireul	0.07	0.00	0.02	0.04	0.07	0.02	0.04
Pseudorasbora parva	0.09	0.01	0.10	0.13	0.00	0.23	0.22
Rhodeus amarus	0.06	0.07	0.03	0.11	0.19	0.17	0.02
Rutilus ohridanus	0.39	0.09	0.46	0.42	0.12	0.07	0.02
Salmo ohridana	0.00	0.00	0.00	0.00	0.08	0.03	0.09
Scardinius knezevici	0.07	0.01	0.17	0.05	0.00	0.03	0.00
Squalius squalus	0.01	0.01	0.05	0.01	0.00	0.00	0.00

ind./m2

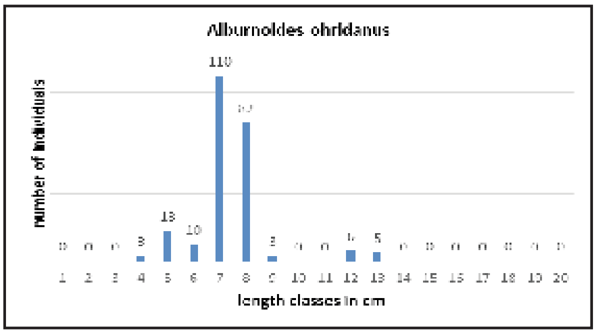
Figure 35. CPUE expressed in number of individuals/m^2 in percentage per species of total catch per depth strata (NPUE) at SB 5 locality

Similar situation like for the biomass distribution is recorded also for the species distribution per number and per depth strata. It is worth to mention that in the depth till 6 m significant dominance of the Ohrid minnow – Pachyhilon – in number is quite evident.

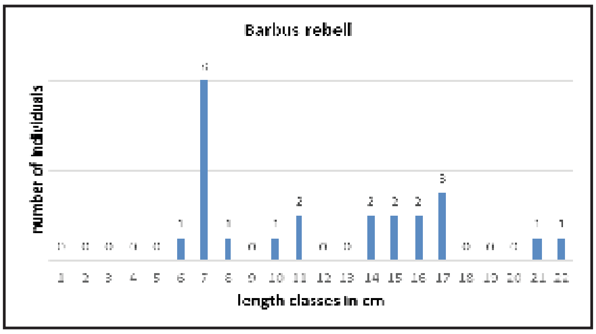
The length frequency distributions of the fish species caught during the survey at the SB 5 are presented in the following figure (36; a-n).



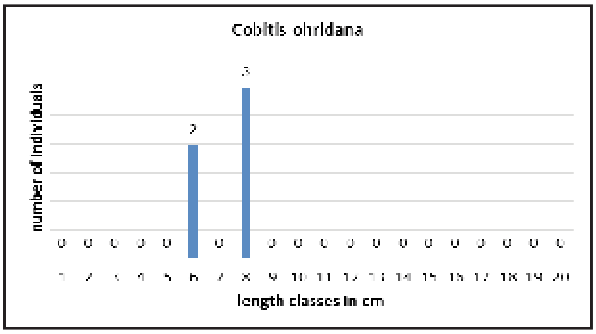
a)



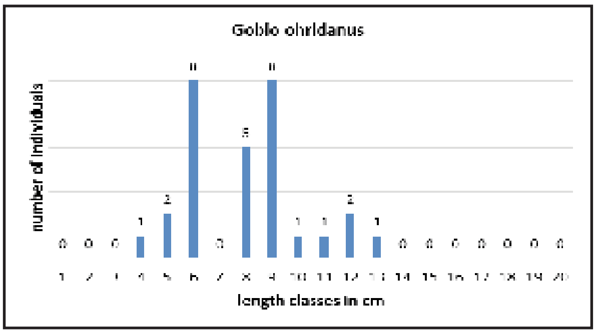
b)



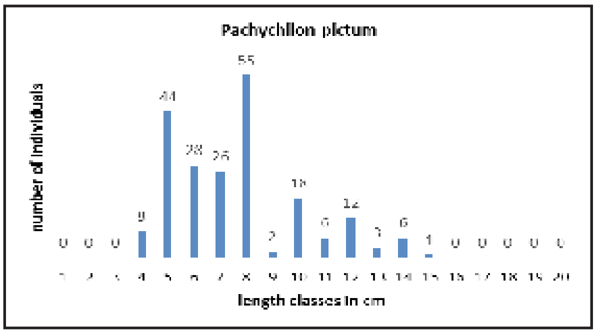
c)



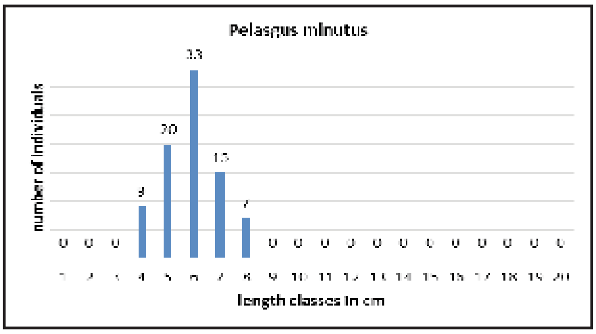
d)



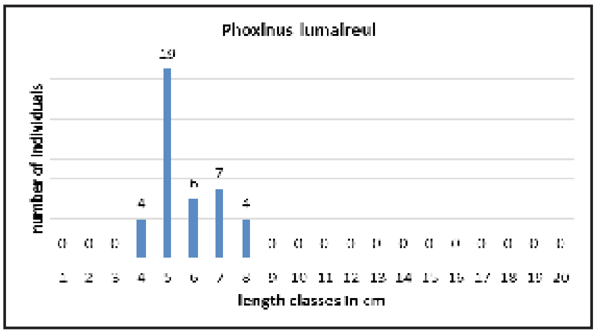
e)



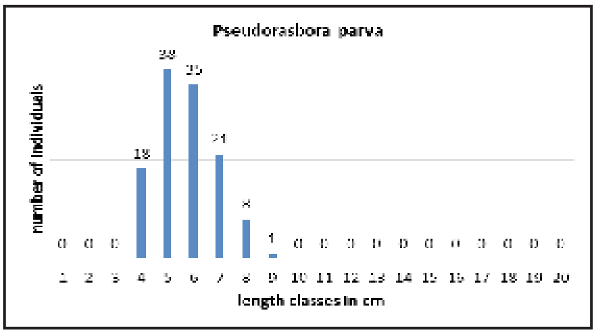
f)



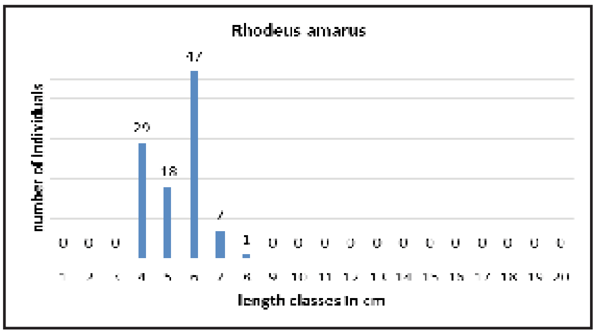
g)



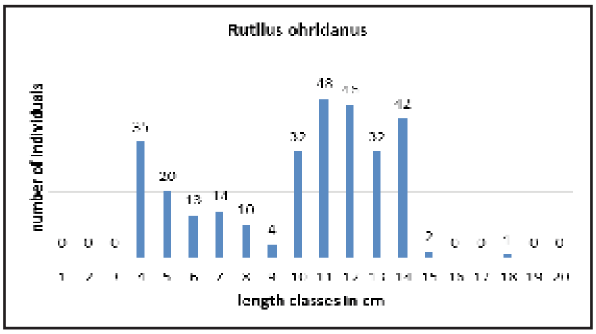
h)



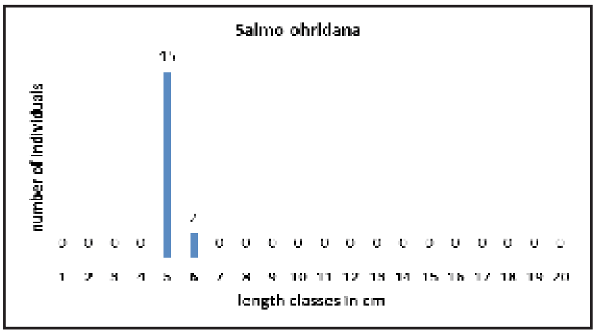
i)



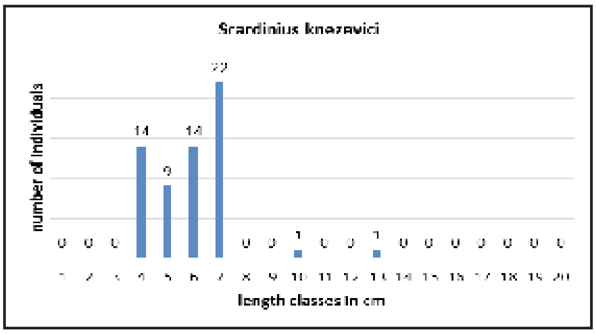
j)



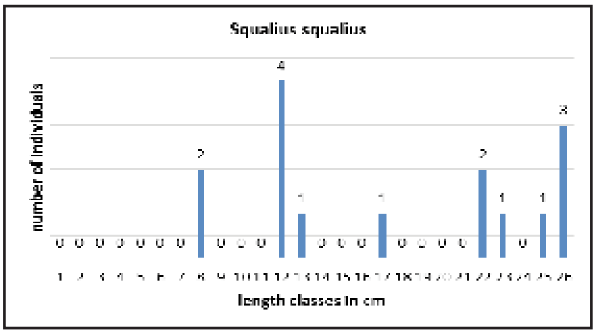
k)



l)



m)



n)

Figure 36. Length classes frequency per species for the SB5 Lin – Bakalice (a-n)

The composition of the species length (age) classes distribution is relatively more comprehensive in the following manner: dominancy of the following species by their respectful number in classes – roach 14, bleak 13, barbell 11, Ohrid minnow (Pachychilon) 11 and the chub with 8 classes.

From other hand, such kind of variety present species reflects the characteristics of the sub basin.

4.3.1.6. SB6 – Hudunisht

The sampling at SB6 took time within two campaigns. This sub basin from other hand is representing one of the most productive fishing areas of the lake. This SB, along the shore and towards the deep, differs very much in microhabitat's conditions.

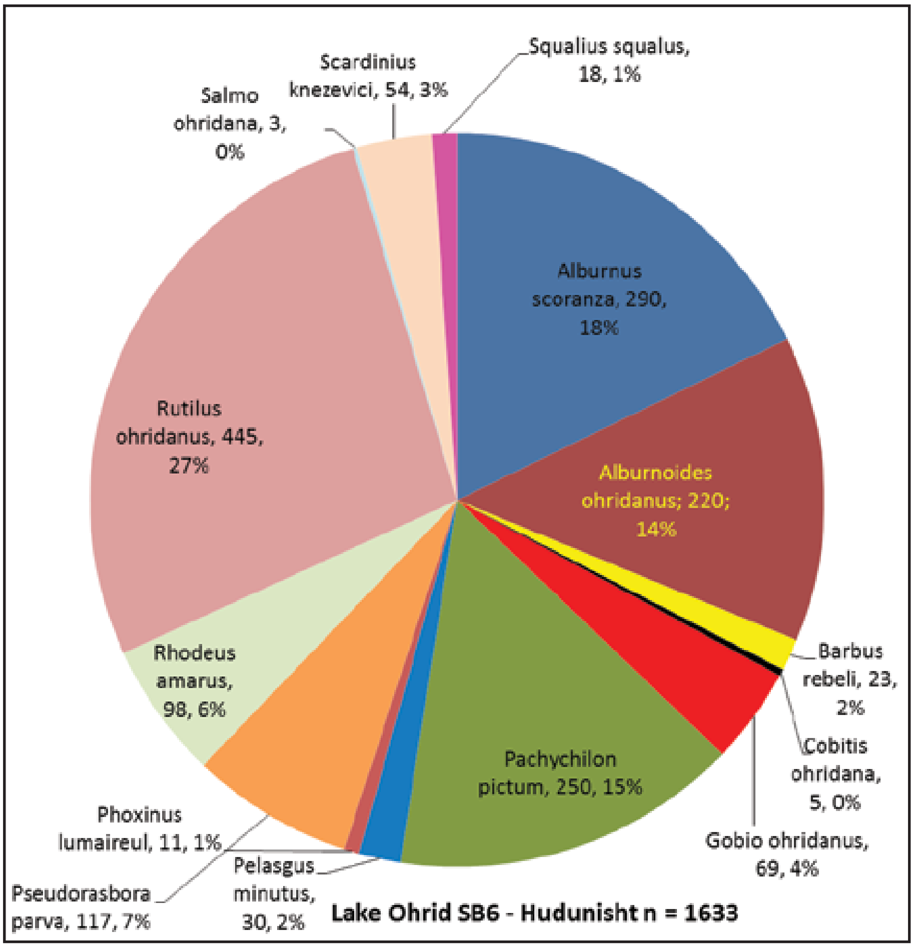
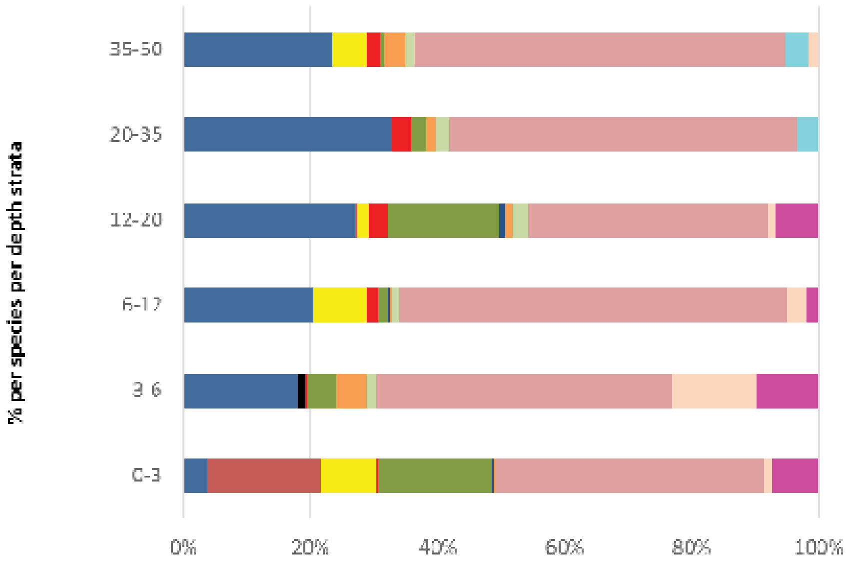


Figure 37. Relative fish species composition in total catch at SB6 in the sampling campaign November 2013

From the sampling at sub basin 6, the results showed the dominance of low valued commercial species roach and bleak or noncommercial species minnow – pachychilon and spirlin. Although, the fishing was performed even at rather deep strata, the catch of the commercially valued species was negligible - 3 individuals of the endemic Ohrid belvica. At this sub basin totally 14 fish species were caught.

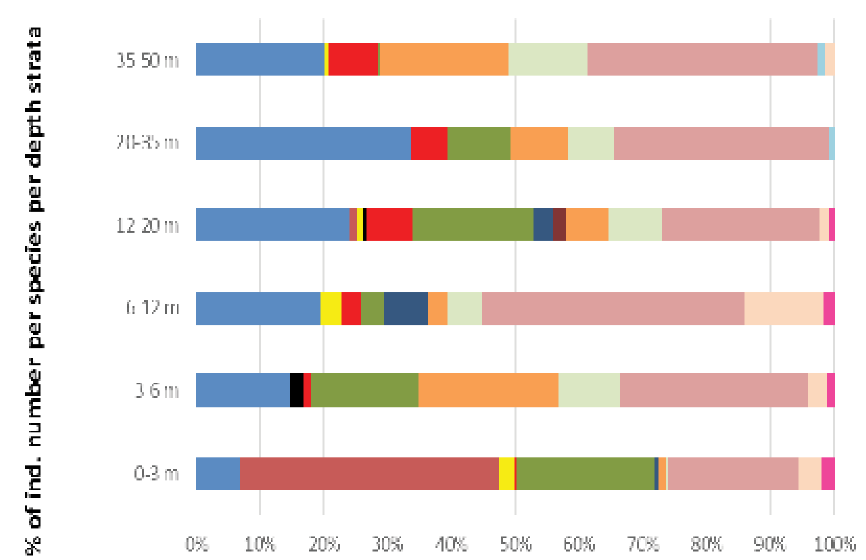


	0-3	3-6	6-12	12-20	20-35	35-50
Alburnus scoranza	0.38	3.20	2.61	4.17	3.85	2.83
Alburnoides ohridanus	4.52	0.00	0.00	0.05	0.00	0.00
Barbus rebeli	2.25	0.00	1.10	0.28	0.00	0.63
Cobitis ohridana	0.00	0.23	0.00	0.02	0.00	0.00
Gobio ohridanus	0.05	0.04	0.20	0.44	0.36	0.26
Pachychilon pictum	4.59	0.80	0.22	2.71	0.30	0.09
Pelasgus minutus	0.04	0.00	0.05	0.11	0.00	0.00
Phoxinus lumaireul	0.00	0.00	0.00	0.03	0.00	0.00
Pseudorasbora parva	0.06	0.86	0.04	0.17	0.20	0.39
Rhodeus amarus	0.01	0.27	0.14	0.38	0.23	0.17
Rutilus ohridanus	10.92	8.31	7.80	5.80	6.15	7.07
Salmo ohridana	0.00	0.00	0.00	0.00	0.40	0.45
Scardinius knezevici	0.32	2.32	0.39	0.20	0.00	0.19
Squalius squalus	1.80	1.76	0.25	1.04	0.00	0.00

g/m²

Figure 38. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 6 locality

The biomass distribution in this case is shared between the roach and bleak with significant prevalence of the roach. Third by dominancy is the Ohrid minnow (Pachychilon), and the chub has also significant part of the biomass distribution per some of the depth strata regarding the sizes of the caught specimens.



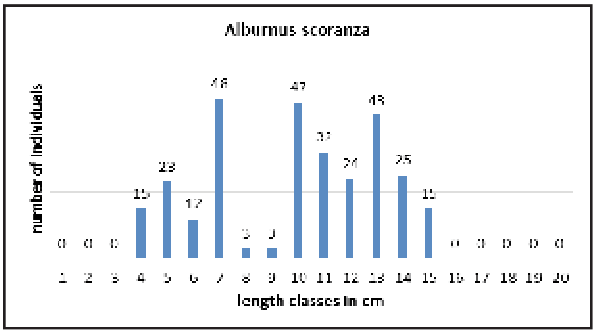
	0-3 m	3-6 m	6-12 m	12-20 m	20-35 m	35-50 m
Alburnus scoranza	0.20	0.31	0.18	0.36	0.30	0.22
Alburnoides ohridanus	1.19	0.00	0.00	0.02	0.00	0.00
Barbus rebeli	0.07	0.00	0.03	0.01	0.00	0.01
Cobitis ohridana	0.00	0.04	0.00	0.01	0.00	0.00
Gobio ohridanus	0.01	0.02	0.03	0.11	0.05	0.08
Pachychilon pictum	0.64	0.36	0.03	0.28	0.09	0.01
Pelasgus minutus	0.02	0.00	0.06	0.04	0.00	0.00
Phoxinus lumaireul	0.00	0.00	0.00	0.03	0.00	0.00
Pseudorasbora parva	0.03	0.47	0.03	0.10	0.08	0.22
Rhodeus amarus	0.01	0.20	0.05	0.13	0.07	0.13
Rutilus ohridanus	0.60	0.62	0.37	0.36	0.30	0.39
Salmo ohridana	0.00	0.00	0.00	0.00	0.01	0.01
Scardinius knezevici	0.11	0.07	0.11	0.02	0.00	0.02
Squalius squalus	0.06	0.02	0.02	0.01	0.00	0.00

ind./m2

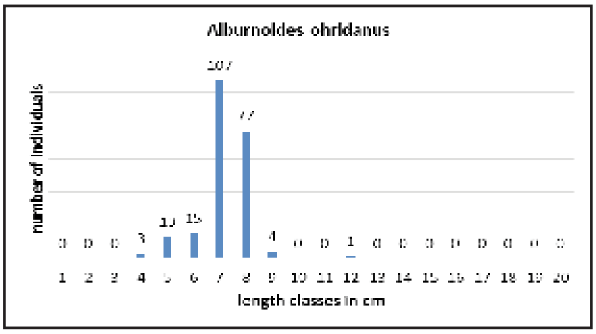
Figure 39. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 6 locality

Also at this sub basin 6, the distribution of the individuals per depth strata corresponds more or less with the biomass distribution per species. Exception can be meant for the spirlin in the first depth strata 0-3m, where its dominance is very obvious.

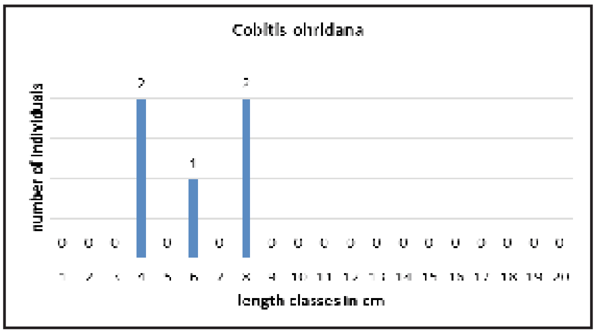
The length frequency distributions of the fish species caught during the survey at the SB 6 are presented in the following figure (40; a-o).


Alburnus scoranza
number of individuals
length classes in cm

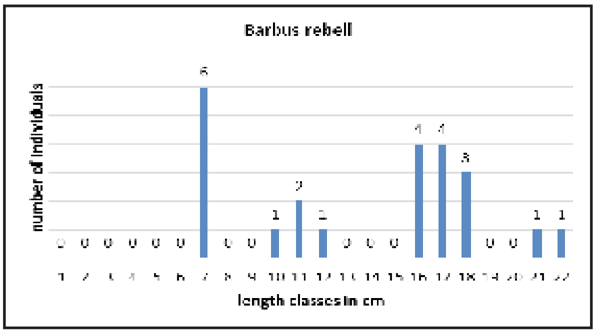
a)


Alburnoides ohridanus
number of individuals
length classes in cm

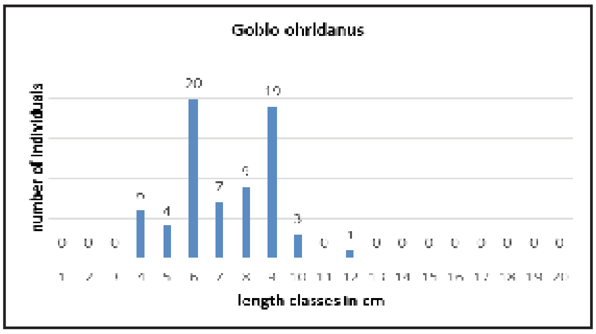
b)


Cobitis ohridana
number of individuals
length classes in cm

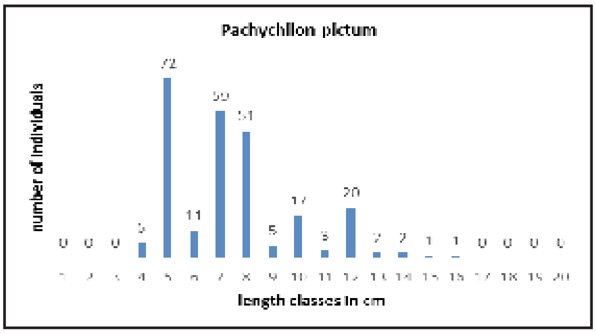
c)


Barbus rebeli
number of individuals
length classes in cm

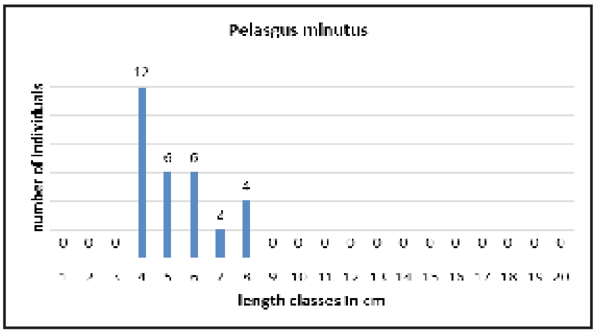
d)


Gobio ohridanus
number of individuals
length classes in cm

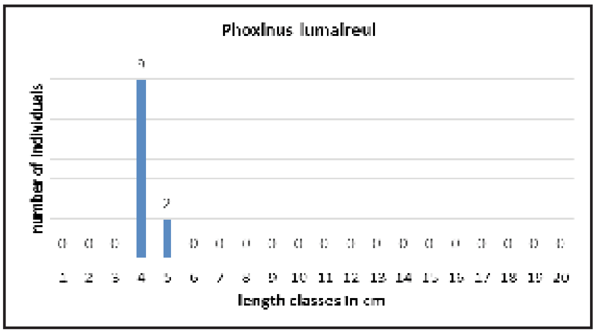
f)


Pachychilon pictum
number of individuals
length classes in cm

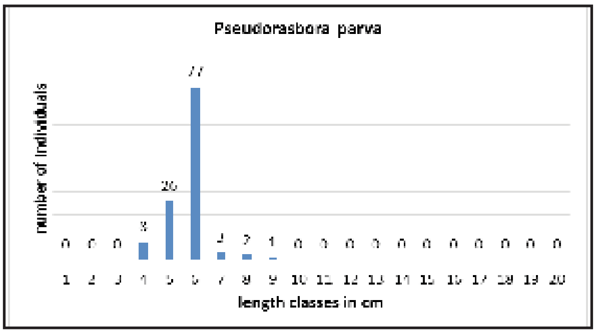
g)


Pelasgus minutus
number of individuals
length classes in cm

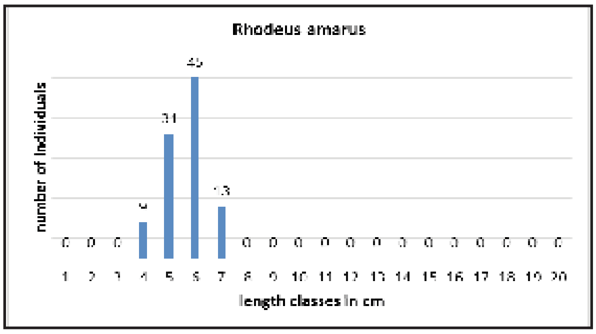
h)


Phoxinus lumaireul
number of individuals
length classes in cm

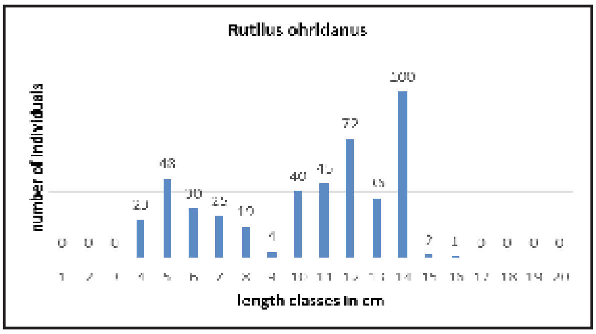
i)



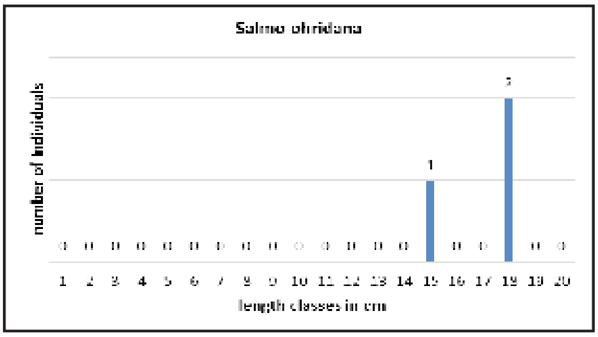
j)



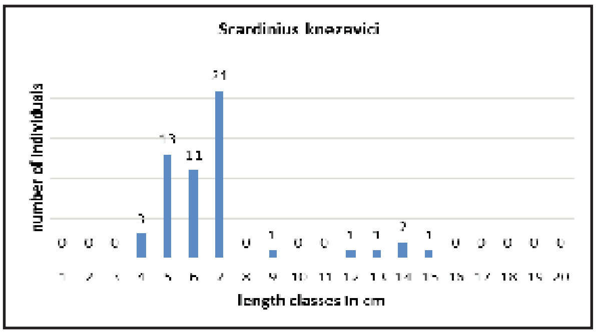
k)



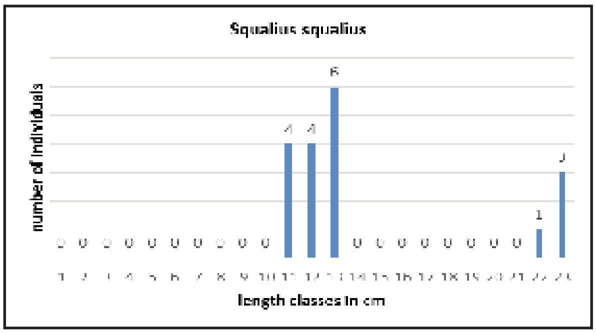
l)



m)



n)



o)

Figure 40: Length classes frequency per species for the SB5 Hudunist (a-n)

From the figures above, it can be stated that the dominance of different length classes of relevantly significant species is as follows: roach and Ohrid minnow with 13 length classes, the bleak with 12 and barbell with 9 classes.

4.3.1.7. SB7 – Tushemisht

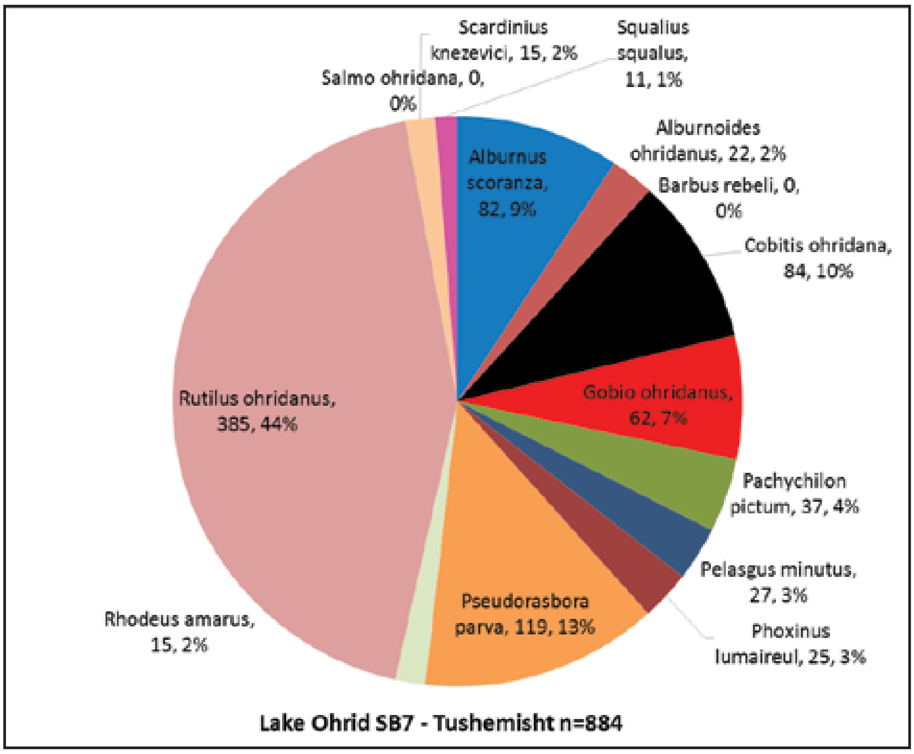
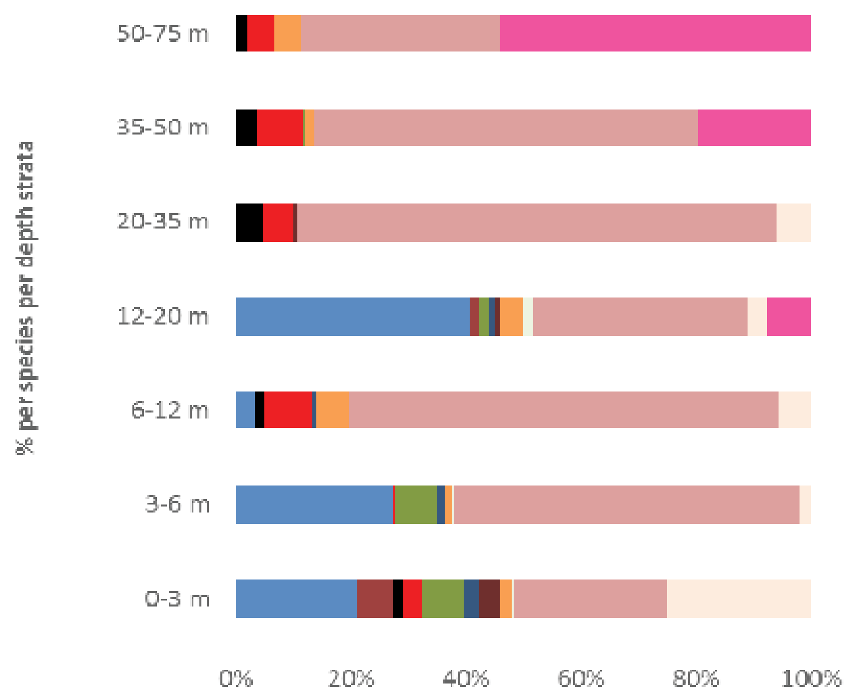


Figure 41. Relative fish species composition in total catch at SB7 in the sampling campaign November 2013

Only 12 species were caught in Tushemisht area. Barbell and belvica were not present in the nets. Also some species such as spirlin, rudd, and chub are present in only some depths and not in the others. Unlike in the other sub-basins, the dominant species is clearly roach in almost all depths while bleak presence here is much more reduced.

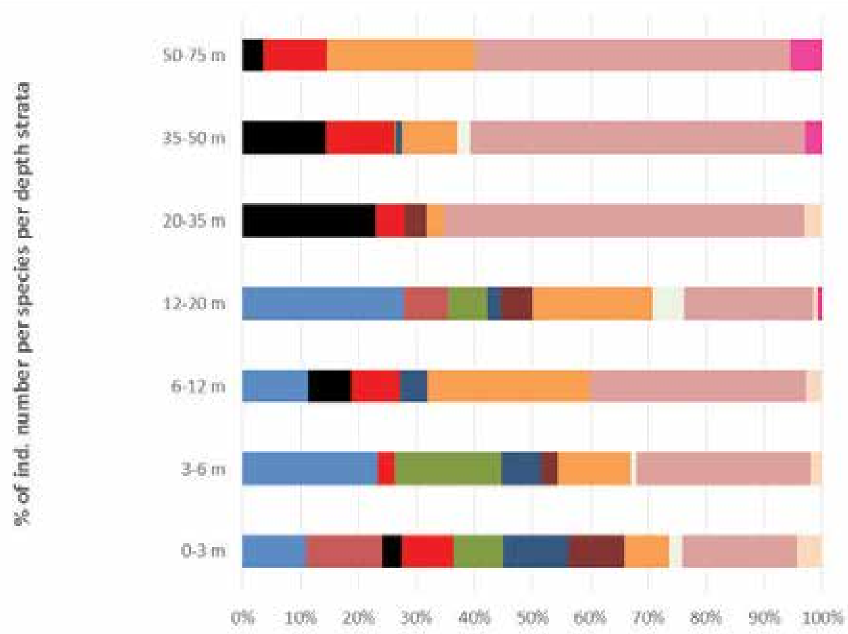


	0-3 m	3-6 m	6-12 m	12-20 m	20-35 m	35-50 m	50-75 m
Alburnus scoranza	0.72	2.70	0.28	6.61	0.00	0.00	0.00
Alburnoides ohridanus	0.22	0.00	0.00	0.28	0.00	0.00	0.00
Cobitis ohridana	0.05	0.00	0.16	0.00	0.49	0.33	0.17
Gobio ohridanus	0.13	0.04	0.76	0.00	0.52	0.69	0.48
Pachychilon pictum	0.25	0.70	0.00	0.27	0.00	0.01	0.00
Pelasgus minutus	0.09	0.14	0.07	0.15	0.00	0.01	0.00
Phoxinus lumaireul	0.12	0.02	0.00	0.14	0.09	0.00	0.00
Pseudorasbora parva	0.07	0.12	0.47	0.66	0.02	0.12	0.41
Rhodeus amarus	0.01	0.03	0.00	0.25	0.00	0.02	0.00
Rutilus ohridanus	0.91	5.93	6.75	6.07	8.57	5.80	3.33
Scardinius knezevici	0.87	0.20	0.50	0.56	0.60	0.00	0.00
Squalius squalus	0.00	0.00	0.00	1.24	0.00	1.72	5.17

g/m²

Figure 42. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 7 locality

The values of CPUE expressed in biomass per square meter of net here are smaller compared to other sub-basins. There is a dominance of the roach that is quite marked in all depths. Bleak has a good presence until 20 m depths and then disappears completely. Except for roach, most values of BPUE do not go beyond 1 g/m² which is quite low compared to other sub basins where values above 2 g/m² are common for several species. It should be noted that in the catch for high depths (> 35 m) the presence of chub in terms of BPUE is quite relevant.

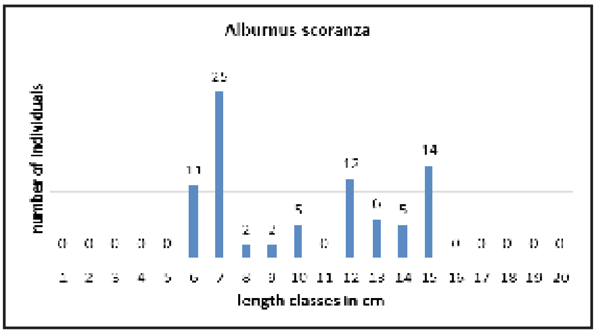


	0-3 m	3-6 m	6-12 m	12-20 m	20-35 m	35-50 m	50-75 m
Alburnus scoranza	0.06	0.18	0.09	0.40	0.00	0.00	0.00
Alburnoides ohridanus	0.07	0.00	0.00	0.11	0.00	0.00	0.00
Cobitis ohridana	0.02	0.00	0.06	0.00	0.16	0.13	0.04
Gobio ohridanus	0.04	0.02	0.07	0.00	0.04	0.10	0.13
Pachychilon pictum	0.04	0.14	0.00	0.10	0.00	0.00	0.00
Pelasgus minutus	0.06	0.05	0.04	0.03	0.00	0.01	0.00
Phoxinus lumaireul	0.05	0.02	0.00	0.08	0.03	0.00	0.00
Pseudorasbora parva	0.04	0.10	0.22	0.30	0.02	0.09	0.31
Rhodeus amarus	0.01	0.01	0.00	0.08	0.00	0.02	0.00
Rutilus ohridanus	0.10	0.23	0.30	0.32	0.44	0.51	0.67
Scardinius knezevici	0.02	0.01	0.02	0.01	0.02	0.00	0.00
Squalius squalus	0.00	0.00	0.00	0.01	0.00	0.03	0.07

ind./m²

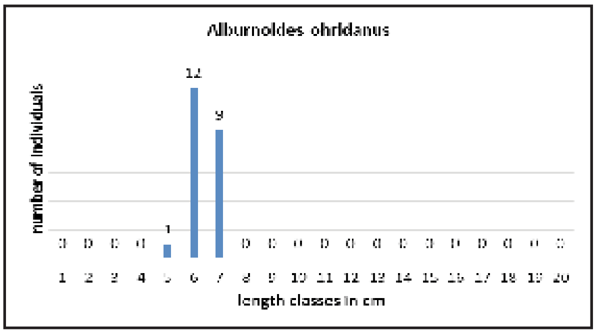
Figure 43. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 7 locality

Also judging from the CPUE expressed as number of individuals per square meter the catch values in Tushemisht sub-basin are at least 30% smaller compared to other areas. The NPUE values shows even a greater dominance of roach in all depths (in > 20 m depths individuals from these species constitute more than 50% of the catch). The only depth where the composition of species is more uniform is the 0-3 m depth where there are at least 5 species that are quite present in the catch. Species like spined loach and stone moroko that were rare in other sub-basins, in Tushemisht were more frequent in several depths.



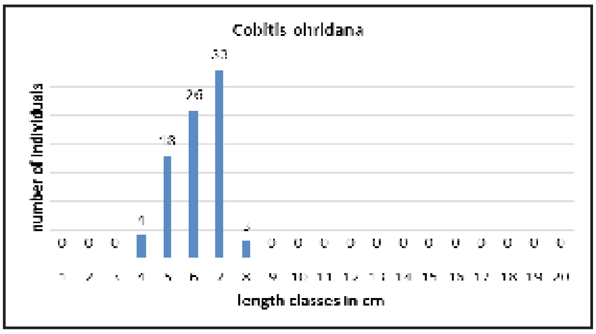
Alburnus scoranza
number of individuals
length classes in cm

a)



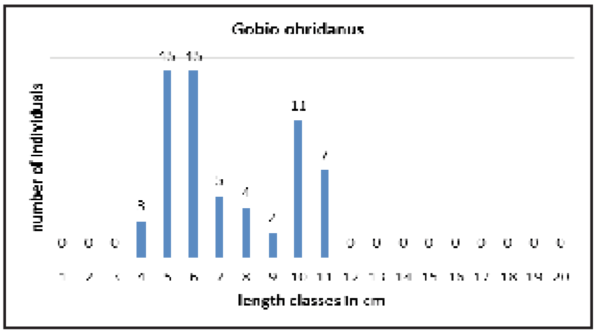
Alburnoides ohridanus
number of individuals
length classes in cm

b)



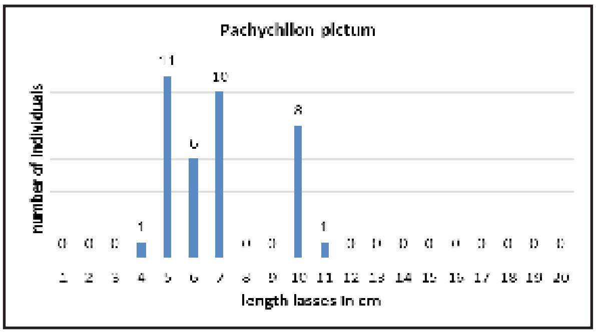
Cobitis ohridana
number of individuals
length classes in cm

c)



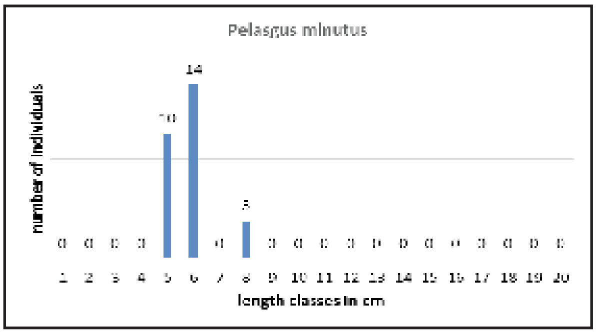
Gobio ohridanus
number of individuals
length classes in cm

d)



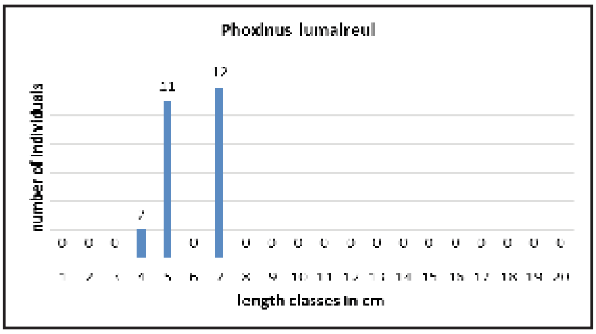
Pachychilon pictum
number of individuals
length lasses in cm

e)



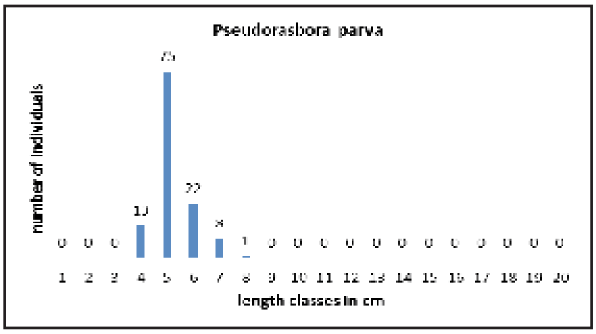
Pelasgus minutus
number of individuals
length classes in cm

f)



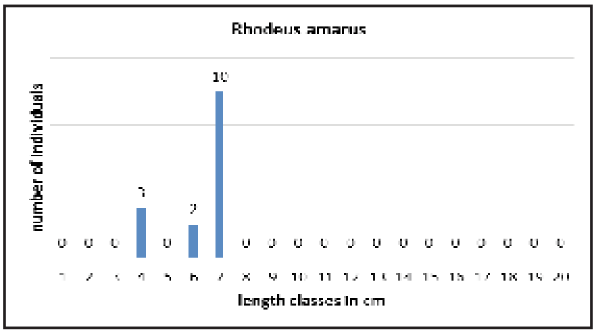
Phoxinus lumaireul
number of individuals
length classes in cm

g)

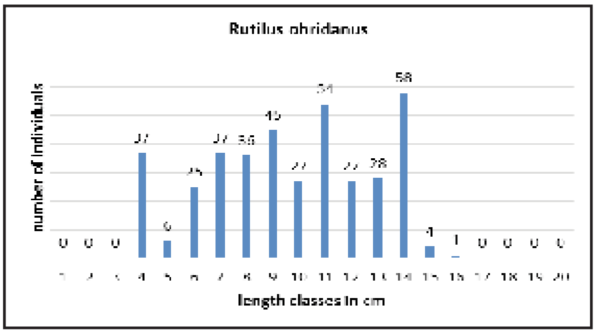


Pseudorasbora parva
number of individuals
length classes in cm

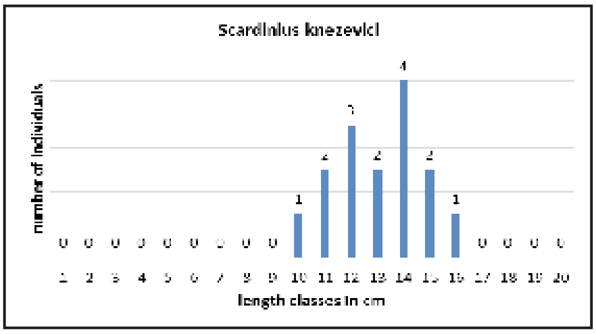
h)



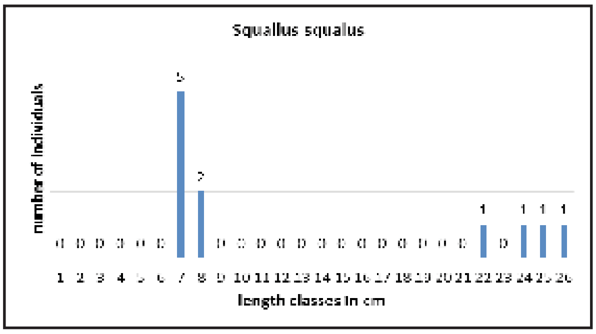
i)



j)



k)



l)

Figure 44. Length classes frequency per species for the SB7 Tushemisht (a-l)

Despite being in fewer numbers, the distribution for bleak and spirlin is similar to other sub-basins. The major frequency of the bleak is in two peaks one at 6-7 cm and one at 12-13 cm, while the majority of the spirlin individuals are in the 6-7 cm length classes. It should be noted that in Tushemist, the roach are somehow evenly distributed in 4-14 cm length classes while in Lin and Hudunisht the 8 and 9 cm classes are almost non-existent.

4.3.1.8. Synthesis for Lake Ohrid

Till this project, never before this kind of multi mesh size gill nets fishing have been performed. Similarities or dissimilarities in terms of species richness based on MMG were also not performed. From the unpublished data (Spirkovski and Ilik-Boeva 2014) mainly the differences in different habitats were related to the spawning grounds of the salmonid species. Previous habitats fish inhabitants' composition was made with gears that are used for the commercial fishery and served like bases for the needs of the relevant ministries and fishing companies. Mainly this experimental fishing was performed in the periods of assembling for spawning or wintering at different localities. The species of non-commercial value were not treated significantly, but like "by catch".

Thus, in this case we can speak only of habitat similarities or dissimilarities tide for the littoral and sub-littoral parts of the lake.

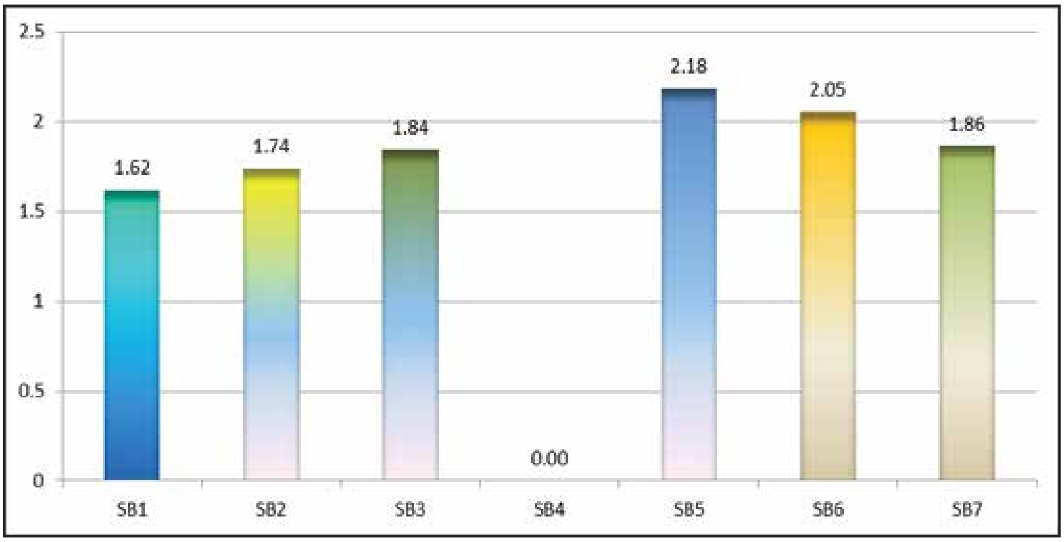


Figure 45. Biodiversity (Shanon-Wiener) index of the seven sampled sub-basins of the water body Lake Ohrid

But, based on the sampling campaign at Lake Ohrid for the MMG in second half of October and first half of November in 2013, the applied Shennon – Wiener index based on biomass/m^2 gives not so big discrepancies among the whole lake. This might be result of the sampling period, the used MMG gear and the weather conditions during the sampling campaign.

Anyway, from Fig.43 it is obvious that the sub basins at the Macedonian part SB1-SB4 unlike those on the Albanian part SB5-SB7 are with lower fish yield. This is very obvious if we have into account the number of fish individuals caught on the Macedonian side – 3215 and 4099 at the Albanian part of the lake. This is very well supported by the fig.44 bellow.

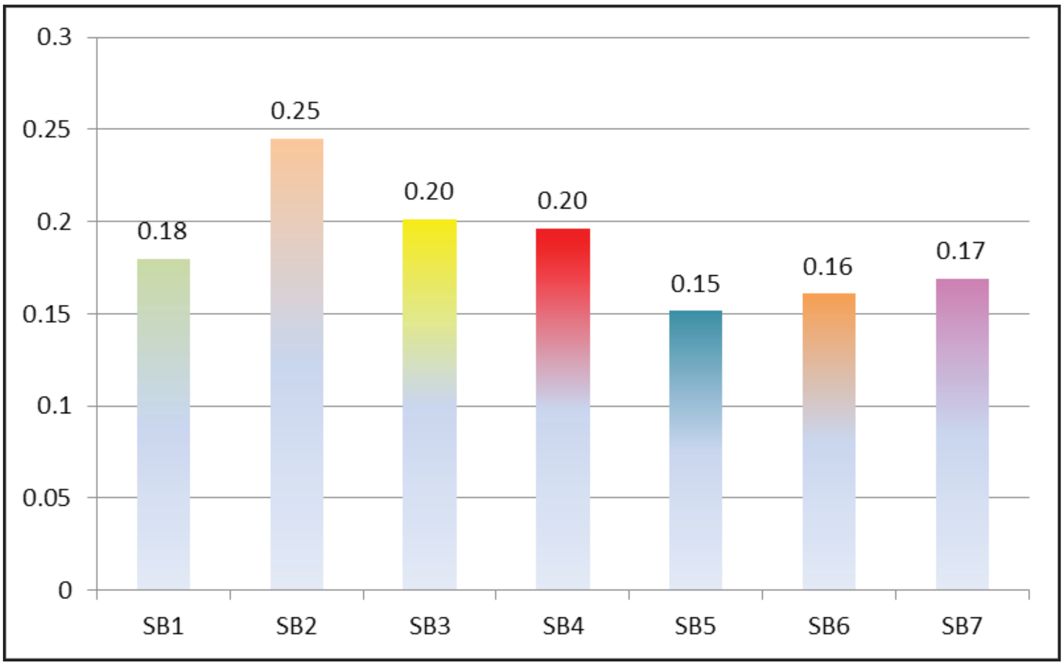


Figure 46. Evenness variation index represented fish species in the seven sampled sub- basins of the water body Lake Ohrid

The evenness or species richness per sub basin expressed in presence of species and their number for a given sub basin, is showing that species richness is reversely from the number of caught individuals versus present species. Thus, the lower the index values are, richer in species the sub basin is. Being represented only with one species the SB4 its involvement in this kind of indexes might be questionable.

4.4. Discussion of results trans-boundary sampling scheme

To adopt the standard EN 14757 from one side and to fulfil the same for large water body as Lake Ohrid is, random sampling with MMGN was performed on seven different (by habitats, wind exposure, water currents, ecological conditions, addition nutrient load and exposure to harmful substances) sub-basins. In total 135 nets were set at both sampling parts of the Lake, 64 in Macedonia and 72 in Albania.

This figure in fact represents the actual (instantaneous – ten days sampling campaign) fish assemblage in October and November 2013. In total, 16 fish species were recorded in the catch. These results which are showing dominance of the native species (see Tab. 1) with more than 84% of the total catch can serve mainly about determining the ecological status of certain habitats and also knowledge of the alien species ecology in relation to manage or design certain strategies for their reduction. From other side, regarding fishery issues (catch quotas, minimum catchable size, types of gears or even simple restricted fish stock assessment) the obtained data are not usable.

Taking Lake Ohrid as one water body, regarding the biomass of fish species, three native (bleak roach and spirlin) and two alien species (pumpkinseed and bitterling) are predominant. On the other side, regarding the number of individuals per square meter of net (NPUE) at SB4 only one species Ohrid gudgeon was caught so it is present with 100 %.

SB1

From the sampling at this sub basin, the results showed species dominance of mostly noncommercial Ohrid spirlin and minnow – pachychilon or low valued commercial species roach and bleak. Although, the fishing was performed even at rather deep strata, the catch of the commercially valued species was negligible - 2 individuals of the endemic Lake Ohrid belvica and 1 carp (Figure 17). At this sub basin totally 14 fish species were caught.

The structure of the bleak and the roach population gives normal size and age distribution for this period of the year. Thus, the roach is represented with 14 length classes and the bleak with 12. Significant to mention is the number of the 10 length classes of the Ohrid barbell. But regarding the older knowledge about its presence at this sub basin from the fishery statistics of the fishing company, it appears that the barbell population is improving at this region.

The most important is that the salmonid species were very insignificantly represented and only Lake Ohrid belvica with 2 specimens, which may be result of relatively unfavorable water temperature, higher than 15 ^{0}C or of the type of the fishing gear used.

SB2

At this sub basin, is obvious the dominance of the roach and other zoobenthofagous species, where amongst the native species, significant presence of the aliens stone moroko and bitterling is evident. At this sub basin totally 13 fish species were caught. Expressed in individuals per species, here apart from the dominancy of the roach, the spine and stone loaches from the natives and stone moroko are relatively high represented. From the length classes of the native fishes dominates the roach with 11 sizes, and followed by the Ohrid gudgeon with eight classes.

SB3

The sampling at this sub basin, the results showed dominance of commercially non valued species (minnows and alien, stone moroko) and whilst the low commercially valued species like bleak and roach represented together with 41% , the more attractive fish were represented only with 6 barbell specimens. All together 12 fish species were caught.

Regarding CPUE for this sub basin, the dominance in biomass is almost equal to the dominance in number represented in the catch. Thus, the roach, also here, is the leading one in almost all presented depth strata. The bleak dominates significantly in the shallower strata 0-6 m. The Ohrid gudgeon is significantly represented in the last two deepest strata.

In terms of number of specimens in this case also the roach is most dominant with 12 length classes, followed with 10 of the Ohrid minnow (*Pachychilon*) and 9 of bleak length classes.

SB4

At the pelagic sub-basin only one species - Ohrid gudgeon with 23 individuals were caught in 10 nets.

SB5

From the sampling at sub basin 5, the results showed the dominance of low valued commercial species bleak and roach or noncommercial species Ohrid minnow-Pachychilon and spirlin. Although, the fishing was performed even at rather deep strata, there was no presence of the commercially valued species. At this sub basin totally 13 fish species were caught.

Unlike the previous described sub basins belonging to the Macedonian part, at this sub basin the rudd (*Scardinius knezevici*) appears in the catches.

The composition of the species length (age) classes distribution is relatively more comprehensive in the following manner: dominancy of the following species by their respectful number in classes – roach 14, bleak 13, barbell 11, Ohrid minnow (Pachychilon) 11 and the chub with 8 classes.

SB6

At this sub-basin dominance of low valued commercial species roach and bleak or noncommercial species minnow – pachychilon and spirlin. Although, the fishing was performed even at rather deep strata, the catch of the commercially valued species was negligible - 3 individuals of the endemic Ohrid belvica. At this sub basin totally 14 fish species were caught.

Here it can be stated that the dominance of different length classes of relevantly significant species is as follows: roach and Ohrid minnow with 13 length classes, the bleak with 12 and barbell with 9 classes.

SB7

Only 12 species were caught in Tushemisht area. Barbell and belvica were not present in the nets. Also some species such as spirlin, rudd, and chub are present in only some depths and not in the others. Unlike in the other sub-basins, the dominant species is clearly roach in almost all depths while bleak presence here is much more reduced.

Till this project, never before this kind of multi mesh size gill nets fishing have been performed. Similarities or dissimilarities in terms of species richness based on MMG were also not performed. From the unpublished data (Spirkovski and Ilik-Boeva 2014) mainly the differences in different habitats were related to the spawning grounds of the salmonid species.

But unlike the expectations that the MMG net fishing will gave some results for the most commercially and endemic species like the Ohrid trout and belvica and the common carp, the sampling campaign didn't have any significant results for them.

Regarding the usage of different diversity indexes isn't fully valid data for characterization of the tested sub basins, especially for Lake Ohrid, where according to Naumovski 1985, the maximum depth is 164 m.

Based on the sampling campaign at Lake Ohrid for the MMG in second half of October and first half of November in 2013, the applied Shennon – Wiener index based on biomass/m^2 gives not so big discrepancies among the whole lake. This might be result of the sampling period, the used MMG gear and the weather conditions during the sampling campaign.

Looking in general from the previous fish and fishery investigations on Lake Ohrid fish fauna in terms of abundance, spawning grounds, fish yield (mainly from reports to the relevant ministries and fishing companies) it can be concluded that using the EN 14757 standard with MMG can not give real picture for such large lake like Ohrid.

But, nevertheless it gave results about the presence and distribution of the alien species (stone moroko and bitterling) while the rest of the previously evidenced aliens didn't appear in the catch (rainbow trout, pumkinseed, Prussian carp and moskito fish).

The presence of alien species in terms of number of individuals of bitterling and stone moroko the role as aliens in relation to natives in Lake Ohrid is poorly understood. The WFD status class has been discussed with a number of people and it seems to be general agreement that the presence of one or more established alien species is certainly not 'natural' and is likely to affect the WFD status class in a detrimental way.

Also, the main commercially important fish like Ohrid trout and belvica and the common carp were negligible in the catch, even without any specimen of Ohrid trout. This could perhaps not be a case if the planned pelagic MMG sampling was performed, so we would have totally different picture about Lake Ohrid fish population.

Another important thing is the total absence of the Ohrid nase (*Chondrostoma ohridanus*) in the catch. The reduced population of this fish has been recorded before but the opinion was that the fishers are not using proper gear for it to be caught.

From other hand, the presence of the barbell in almost all of the sub basins gives positive signs of its population recovery.

The usage of biodiversity indexes or species richness in the case of Lake Ohrid is far from giving significant data about the sub basins similarities or dissimilarities.

5. Conclusions

As already known and stated in lot of papers scientific and technical, the oligotrophic character of Lake Ohrid (< 8 $mg \cdot m^{-3}$ T P) respectfully has also a relatively low fish production, which varies within the range of 7-12 $kg \cdot ha^{-1}$. Opposite from low fish biomass production as already known in the basic limnological literature it has bigger native fish species richness than those highly productive in biomass.

Although the first written scientific records for Lake Ohrid biodiversity richness with endemic and archaic faunistic species were started with the endemic fish species *Salmo ohridana* 1892 (Lake Ohrid belvica) described by the famous Austrian zoologist Staindachner, further on Lake Ohrid fish fauna wasn't sufficiently investigated.

Most of the investigations of Lake Ohrid fishes were considered to taxonomic and some ecological viewpoints. Until now apart from some sporadic investigations almost no proper fish stock assessment and inventarisation has been made.

From 135 randomly set MMGN in the 2013 sampling campaign 16 fish species (Table 1) were detected in total from which only two alien species, stone moroko - *Pseudorasbora parva* and bitterling – *Rhodeus amarus*. Remaind uncaught are 3 native species: Ohrid trout, eel and nase and the rest are aliens: rainbow trout, pumkinseed, Prussian carp and moskito fish.

The attempts from this CSBL Project were good exercise to implement EN14757 with MMG for the first time on Lake Ohrid. Hence, some interesting results were achieved:

- Using small mesh sizes nets (panels of 5, 6,25 and 8) showed the real significant presence of some of the alien species like stone moroko and bitterling which could not be recorded in the regular fishing gears used by the fishers.
- From other hand despite the depths where the nets were set (till 165 m) and due to their construction it appears that the most dominant fishes in the professional (commercial) fisherman catch like Ohrid trout, Ohrid belvica and carp were almost absent. Thus this EN 14757 method could not be applied for those species as in their abundance, distribution and spatial presence in such large water body. Most probably if we've had the possibility to have and use the other planned fishing gears this situation could be most likely improved.

The main extraction of the fish biomass was and is fishing. Regarding the fishing on Macedonian side from 2004 till 2012 there was total fishing moratorium, whilst in a meanwhile on the Albania side the number of fishermen was increased. Worth to mention here is that there is no reliable fishery statistic for the Albanian part for this period.

The present monitoring survey and data collected serve as a prove for the usefulness of applying standardized lake fish parameters to assess the ecological status of lakes and support the development of integrated monitoring of entire lake. Further to that following European experiences development and setting ecological quality based on different metrics.

Applied protocol and sampling scheme used within CSBL MMGN sampling in future should be widened as in fishing gears and other fish surveillance devices as well for time series.

6. Recommendations

Lake Ohrid is a shared resource, and no action can be taken by one country without impacting the resources and conditions in the other country.

It is of utmost importance to re-establish and to reactivate the bilateral co-management authority ("Lake Ohrid Fisheries Authority or Commission"), which already existed in the previous century to manage the fisheries and related resources. Any other experiences similar to the conditions of Lake Ohrid fish fauna and its ecosystem are welcomed and appreciated. Representatives from national institutions, local authorities, fishermen's organizations, research institutions, civil society etc. are recommended to be considered for membership.

These mix authority (technical and political) could be establish in the frame of *The Agreement between Albanian and Macedonian Governments for the Protection and Sustainable Development of Lake Ohrid and its Watershed*, signed by respective prime ministers some years ago. According to this agreement the riparian countries will take the necessary measures, amongst others, to protect the biodiversity (particularly endemic species), to ensure the sustainable use of natural resources, and to prevent and control the economic activities from seriously damaging (and polluting) the environment. In the light of the above possible measures it is very important that the fishing effort be reduced to conserve biodiversity and to restore the balance of the underwater fauna in order to exploit the available resources in a sustainable manner.

Lake Ohrid fish fauna concerned as a world heritage deserves adequate research resources for its protection and human wellbeing.

Joint Monitoring of fish stock and spawning grounds and habitats is one of necessary important actions

Lake Ohrid Fish stock assessment based on time series using all necessary fishing gears and other surveying technics.

Due to the uncertainty regarding the success of trout stocking it is recommended to conduct marking and tagging experiments and to sample catches for hatchery-reared trout.

Proposed measures and actions stated in the following table are from the Transboundary Fish and Fisheries Management Plan for Prespa Lakes Basin (UNDP, 2012) prepared by PSI Hydrobiological Institute-Ohrid and HYDRA with respect to improvement of fisheries management, protection of biodiversity of fish fauna and lowering pressure on fishes and it is adjusted for implementation at Lake Ohrid.

No	Measures	Actions
1.	Bilateral Fishery Management	- Establishing Joint Lake Ohrid Fishery Commission (JLOFC) - Joint Fishery Master Plan for Lake Ohrid
2.	Monitoring of the water quality, fish stock monitoring	- Reinforcement of the local monitoring stations in both countries in cooperation with scientific institutions and other relevant stakeholders
3.	Joint Monitoring Technical Protocol	- Quality Assurance and Data Acquisition (Designated responsible implementing bodies)
4.	Improved Fish Statistics	- Implementing unique software (Data exchange) - Establishing Fishery Data Base
5.	Fish Stock Assessment	- Integrated actions (open cross border expeditions and surveillances with joint resources) FSA - Revision and relevant changes of the actual Fishing Master Plans for Lake Ohrid and Lake Ohrid Watershed on the Macedonian side.
6.	Physical guarding of the fish stocks	- Establishing national guarding bodies (state and private) - Improving the fishery Inspectorates
7.	Conservation	- Conservation action plans per fish species - Implementing new fishery technics for avoiding by-catch - Stocking program only with autochthonous fish related to specific habitats
7.	Alien fishes combat	- Selective and ameliorative fishing
8.	Fishing limits	- Determining the allowable smallest catchable size per species - Determining the spawning periods and close fishing season per species
9.	Spawning grounds - habitats	- Defining strict natural fish spawning grounds (where any activities without special permission of the national management bodies and JOLFC are allowed) - Improving the conditions of spawning grounds - Ghost nets cleaning and habitat restoration
10.	Catch quotas	- Determining of Annual Total Allowable Fish Catch Quotas (ATAFCQ) per country / per species
11.	Fishing regulations	- Maximum allowed fishing gears and fishing equipment for commercial and recreational fishery
12.	Fish Stocking	- Designing of Joint Fish Stocking Program (JFSP) based on obtained fish spawning individuals from the lakes or streams

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8. Attachments

Annex I. Preliminary fish sampling scheme:

Method	Macedonia											
	Jan.	Feb.	Mar.	Apr.	May	Jun.	July	August	September	Oct.	Nov.	Dec.
Benthic multimesh gillnets European Standard							Veli Dab Daljan Kalishta		Veli Dab Daljan Kalishta			
Pelagic multimesh gillnets European standard								Pelagic 240 m depth				
Fyke Net							Daljan Kalishta		Daljan Kalishta			
Electro-fishing transects									Daljan Kalishta			
Larvae Trap							Veli Dab Daljan Kalishta pelagial		Veli Dab Daljan Kalishta pelagial			
Beach Seine								Daljan Kalishta	Daljan Kalishta			
Catch data												

Method	Albania											
	Jan.	Feb.	Mar.	Apr.	May	Jun.	July	August	September	Oct.	Nov.	Dec.
Benthic multimesh gillnets European Standard							Lin (Pojske) Tushemisht		Lin (Pojske) Tushemisht			
Pelagic multimesh gillnets European standard												
Fyke Net							Pojske Tushemisht		Pojske Tushemisht			
Electro-fishing transects												
Larvae Trap							Lin (Pojske) Tushemisht		Lin (Pojske) Tushemisht			
Beach Seine								Pojske Tushemisht	Pojske Tushemisht			
Catch data												

For, SB2 Daljan, SB3 Kalista, SB5 Pojske and SB7 Tushemisht beside the multimesh gillnets, **fyke nets** with wings of 5 and 10 mm mesh size from knot to knot were planned to be used.

Fish larval traps were planned to be used at SB1 Veli Dab, SB2 Daljan, SB3 Kalsta SB5 Lin and SB7 Tushemisht as littoral sub basins as well as in SB6 Central plate as pelagic sub basin to cover presence of the species recruitment – 0^{+} of cyprinid and salmonid fishes in Lake Ohrid.

Two different mesh sizes (5 and 10 mm from knot to knot) of **beach seine nets** as more harmless fishing gear for the sampled individuals and more productive, were foreseen to cover the circadian peaks of fish species. This fishing gear was planned to be used at SB2 Daljan, SB3 Kalista, SB5 Lin and SB7 Tushemisht in two consecutive months.

Annex II. **Lake Ohrid points of sampling and additional sampling data**

LAKE OHRID SUB BASINS POINTS OF SAMPLING (POS) AT MACEDONIAN PART

SUB BASIN	NET NUMBER	SAMPLING DATE	STRATA (m)	ACTUAL DEPTH (m)	LONGITUDE	LATITUDE
SB1 VELI DAB	1	19.10.2013	0 - 3	1.0 - 3.5	E 20° 47´ 941	N 40° 59´ 250
	2		0 - 3	1.5 - 3.5	E 20° 47´ 903	N 40° 59´ 212
	3		3 - 6	5.0 - 5.0	E 20° 47´ 964	N 40° 59´ 304
	4		6 - 12	6.5 - 6.5	E 20° 47´ 946	N 40° 59´ 367
	5		6 - 12	6.5 - 13.0	E 20° 47´ 953	N 40° 59´ 408
	6		6 - 12	6.0 - 15.0	E 20° 47´ 939	N 40° 59´ 464
	7	04.11.2013	3 - 6	3.4 - 3.4	E 20° 48´ 026	N 40° 59´ 950
	8		3 - 6	3.5 - 10	E 20° 47´ 993	N 40° 59´ 937
	9		6 - 12	6.0 - 14.0	E 20° 47´ 886	N 40° 59´ 968
	10		20 - 35	18.0 - 40.0	E 20° 47´ 966	N 41° 00´ 028
	11		12 - 20	13.0 - 15.0	E 20° 48´ 061	N 41° 00´ 075
	12		20 - 35	17.0 - 35.0	E 20° 48´ 108	N 41° 00´ 121
	13		20 - 35	23.0 - 23.0	E 20° 48´ 132	N 41° 00´ 968
	14		3 - 6	3.0 - 6.0	E 20° 48´ 207	N 41° 00´ 308
	15		12 - 20	12.0 - 15.0	E 20° 48´ 190	N 41° 00´ 339
	16		12 - 20	12.0 - 20.0	E 20° 48´ 061	N 41° 00´ 397
SB2 ANDON DUKOV	17	08.11.2012	3 - 6	3.5 - 6.3	E 20° 45´ 568	N 41° 07´ 933
	18		0 - 3	1.5 - 1.9	E 20° 45´ 561	N 41° 08´ 030
	19		3 - 6	3.5 - 4.8	E 20° 45´ 472	N 41° 08´ 043
	20		6 - 12	6.5 - 14.0	E 20° 45´ 385	N 41° 08´ 065
	21		20 - 35	16.0 - 28.0	E 20° 45´ 345	N 41° 08´ 045
	22		20 - 35	22.0 - 25.0	E 20° 45´ 214	N 41° 08´ 171
	23		35 - 50	40.0 - 48.0	E 20° 45´ 124	N 41° 08´ 175
	24		6 - 12	7.2 - 8.2	E 20° 45´ 195	N 41° 08´ 294
	25		6 - 12	9.0 - 13.0	E 20° 45´ 124	N 41° 08´ 283
	26		21 - 35	20.0 - 22.0	E 20° 45´ 134	N 41° 08´ 273
	27	11.11.2013	12 - 20	16.0 - 16.0	E 20° 44´ 392	N 41° 09´ 197
	28		12 - 20	17.0 - 19.0	E 20° 44´ 392	N 41° 09´ 200
	29		12 - 20	19.0 - 18.0	E 20° 44´ 411	N 41° 09´ 212
	30		6 - 12	13.0 - 10.0	E 20° 44´ 165	N 41° 09´ 119
	31		0 - 3	1.0 - 3.0	E 20° 44´ 452	N 41° 09´ 348
	32		12 - 20	18.0 - 28.0	E 20° 44´ 328	N 41° 09´ 077
	33		35 - 50	33.0 - 43.0	E 20° 44´ 536	N 41° 08´ 553
	34		3 - 6	5.0 - 5.3	E 20° 44´ 489	N 41° 09´ 239
	35		35 - 50	45.0 - 57.0	E 20° 44´ 307	N 41° 08´ 558
	36		50 - 75	68.0 - 75.0	E 20° 44´ 106	N 41° 08´ 469
SB4 CENTRAL PLATE	37	12.11.2013	120 - 125	123.0 - 124.0	E 20° 43´ 576	N 41° 08´ 273
	38		120 - 125	123.0 - 124.0	E 20° 43´ 500	N 41° 07´ 450
	39		130 - 135	133.0 - 134.0	E 20° 43´ 350	N 41° 07´ 600
	40		130 - 135	133.0 - 134.0	E 20° 43´ 180	N 41° 07´ 725
	41		130 - 135	133.0 - 134.0	E 20° 42´ 950	N 41° 07´ 515
	42		120 - 125	123.0 - 124.0	E 20° 42´ 870	N 41° 07´ 400
	43		120 - 125	123.0 - 124.0	E 20° 42´ 525	N 41° 07´ 135
	44		165	167.0 - 163.0	E 20° 42´ 194	N 41° 06´ 527
	45		165	167.0 - 163.0	E 20° 42´ 010	N 41° 06´ 300
	46		165	167.0 - 163.0	E 20° 41´ 920	N 41° 06´ 229

SB3 RADOZDA	47	05.11.2013	0 - 3	2.5 - 3.5	E 20° 38´ 214	N 41° 05´ 530
	48		3 - 6	3.7 - 4.2	E 20° 38´ 107	N 41° 05´ 573
	49		6 - 12	6.0 - 14.0	E 20° 38´ 107	N 41° 05´ 370
	50		12 - 20	18.0 - 22.0	E 20° 38´ 231	N 41° 05´ 347
	51		20 - 35	25.0 - 28.0	E 20° 38´ 320	N 41° 05´ 414
	52		20 - 35	30.0 - 30.0	E 20° 38´ 198	N 41° 05´ 548
	53		35 - 50	39.0 - 42.0	E 20° 38´ 265	N 41° 05´ 565
	54		3 - 6	5.0 - 6.0	E 20° 38´ 150	N 41° 05´ 633
	55		3 - 6	5.0 - 5.5	E 20° 38´ 190	N 41° 05´ 684
	56		35 - 50	50.0 - 50.0	E 20° 38´ 409	N 41° 05´ 634
	57	06.11.2013	0 - 3	2.2 - 2.3	E 20° 38´ 836	N 41° 05´ 132
	58		0 - 3	2.5 - 3.2	E 20° 38´ 215	N 41° 05´ 736
	59		12 - 20	7.6 - 22.0	E 20° 38´ 303	N 41° 05´ 747
	60		20 - 35	30.0 - 43.0	E 20° 38´ 340	N 41° 05´ 654
	61		3 - 6	5.4 - 5.0	E 20° 38´ 251	N 41° 05´ 825
	62		6 - 12	3.8 - 11.0	E 20° 38´ 280	N 41° 05´ 883
	63		6 - 12	7.5 - 14	E 20° 38´ 353	N 41° 05´ 831
PELAGIC NET	64	19.10.2013	0 – 6 surface	0.0 – 6.0	E 20° 47´ 754	N 40° 59´ 366

LAKE OHRID SUB BASINS POINTS OF SAMPLING (POS) AT ALBANIAN PART

SUB BASIN	NET NUMBER	STRATA (m)	LONGITUDE	LATITUDE
SB5 LIN	1	0 - 3	E 20° 38´ 450	N 41° 03´ 020
	2	0 - 3	E 20° 38´ 465	N 41° 03´ 030
	3	3 - 6	E 20° 38´ 463	N 41° 03´ 035
	4	3 - 6	E 20° 38´ 475	N 41° 03´ 031
	5	>35	E 20° 38´ 473	N 41° 03´ 039
	6	>35	E 20° 38´ 484	N 41° 03´ 043
	7	20 - 35	E 20° 38´ 456	N 41° 03’ 022
	8	12 - 20	E 20° 38´ 471	N 41° 03´ 028
	9	6 - 12	E 20° 38´ 454	N 41° 03´ 024
	10	3 - 6	E 20° 38´ 480	N 41° 03´ 029
	11	0 - 3	E 20° 38´ 467	N 41° 03´ 021
	12	0 - 3	E 20° 38´ 469	N 41° 03´ 016
	13	0 - 3	E 20° 38´ 452	N 41° 03´ 012
	14	0 - 3	E 20° 38´ 455	N 41° 03´ 018
	15	6 -12	E 20° 38´ 482	N 41° 03´ 010
	16	6 -12	E 20° 38´ 451	N 41° 03´ 008
	17	6 - 12	E 20° 38´ 568	N 41° 03´ 014
	18	20 - 35	E 20° 38´ 561	N 41° 03´ 030
	19	>35	E 20° 38´ 472	N 41° 03´ 043
	20	>35	E 20° 38´ 485	N 41° 03´ 065
	21	>35	E 20° 38´ 445	N 41° 03´ 045
	22	12 - 20	E 20° 38´ 314	N 41° 03´ 038
	23	12 - 20	E 20° 38´ 324	N 41° 03´ 036
	24	12 - 20	E 20° 38´ 495	N 41° 03´ 032

SB6 HUDENISHT	1	0 - 3	E 20° 38´ 152	N 40° 59´ 501
	2	0 - 3	E 20° 38´ 154	N 40° 59´ 503
	3	0 - 3	E 20° 38´ 158	N 40° 59´ 504
	4	0 - 3	E 20° 38´ 163	N 40° 59´ 505
	5	3 - 6	E 20° 38´ 165	N 40° 59´ 507
	6	6 - 12	E 20° 38´ 166	N 40° 59´ 511
	7	6 - 12	E 20° 38´ 171	N 40° 59´ 515
	8	6 - 12	E 20° 38´ 172	N 40° 59´ 512
	9	6 - 12	E 20° 38´ 173	N 40° 59´ 506
	10	12 - 20	E 20° 38´ 175	N 40° 59´ 513
	11	12 - 20	E 20° 38´ 177	N 40° 59´ 508
	12	12 - 20	E 20° 38´ 178	N 40° 59´ 516
	13	12 - 20	E 20° 38´ 179	N 40° 59´ 512
	14	12 - 20	E 20° 38´ 174	N 40° 59´ 518
	15	12 - 20	E 20° 38´ 176	N 40° 59´ 510
	16	12 - 20	E 20° 38´ 181	N 40° 59´ 518
	17	12 - 20	E 20° 38´ 179	N 40° 59´ 516
	18	20 - 35	E 20° 38´ 180	N 40° 59´ 503
	19	20 - 35	E 20° 38´ 181	N 40° 59´ 504
	20	20 - 35	E 20° 38´ 183	N 40° 59´ 506
	21	>35	E 20° 38´ 185	N 40° 59´ 499
	22	>35	E 20° 38´ 187	N 40° 59´ 500
	23	>35	E 20° 38´ 190	N 40° 59´ 501
	24	>35	E 20° 38´ 193	N 40° 59´ 498
SB7 TUSHEMISHT	1	0 - 3	E 20° 43´ 207	N 40° 54´ 164
	2	0 - 3	E 20° 43´ 204	N 40° 54´ 167
	3	0 - 3	E 20° 43´ 202	N 40° 54´ 170
	4	0 - 3	E 20° 43´ 198	N 40° 54´ 178
	5	3 - 6	E 20° 43´ 195	N 40° 54´ 180
	6	3 - 6	E 20° 43´ 193	N 40° 54´ 183
	7	3 - 6	E 20° 43´ 190	N 40° 54´ 187
	8	6 - 12	E 20° 43´ 185	N 40° 54´ 190
	9	6 - 12	E 20° 43´ 181	N 40° 54´ 193
	10	6 - 12	E 20° 43´ 180	N 40° 54´ 198
	11	12 - 20	E 20° 43´ 177	N 40° 54´ 195
	12	12 - 20	E 20° 43´ 176	N 40° 54´ 202
	13	20 - 35	E 20° 43´ 175	N 40° 54´ 204
	14	20 - 35	E 20° 43´ 174	N 40° 54´ 205
	15	20 - 35	E 20° 43´ 172	N 40° 54´ 203
	16	20 - 35	E 20° 43´ 170	N 40° 54´ 210
	17	20 - 35	E 20° 43´ 173	N 40° 54´ 206
	18	>35	E 20° 43´ 145	N 40° 54´ 211
	19	>35	E 20° 43´ 141	N 40° 54´ 215
	20	>35	E 20° 43´ 137	N 40° 54´ 208
	21	>35	E 20° 43´ 132	N 40° 54´ 214
	22	>35	E 20° 43´ 130	N 40° 54´ 218
	23	>35	E 20° 43´ 128	N 40° 54´ 216
	24	>35	E 20° 43´ 125	N 40° 54´ 210

LAKE OHRID DATES OF SAMPLING (POS) AT ALBANIAN PART

Nr	LIN	Nr	HUDENISHT	Nr	TUSHEMISHT
1	0-3 m/6.11.2013	25	0-3 m/5.11.2013	49	>35 m/8.11.2013
2	0-3 m/6.11.2013	26	0-3 m/5.11.2013	50	>35 m/8.11.2013
3	3-6 m/6.11.2013	27	6-12 m/5.11.2013	51	>35 m/8.11.2013
4	3-6 m/6.11.2013	28	6-12 m/5.11.2013	52	20-35 m/8.11.2013
5	>35 m/8.11.2013	29	6-12 m/5.11.2013	53	20-35 m/8.11.2013
6	>35 m/8.11.2013	30	6-12 m/5.11.2013	54	20-35 m/8.11.2013
7	20-35 m/8.11.2013	31	12-20 m/5.11.2013	55	6-12 m/8.11.2013
8	12-20 m/8.11.2013	32	12-20 m/5.11.2013	56	0-3 m/8.11.2013
9	6-12 m/8.11.2013	33	20-35 m/5.11.2013	57	0-3 m/9.11.2013
10	3-6 m/8.11.2013	34	20-35 m/5.11.2013	58	0-3 m/9.11.2013
11	0-3 m/8.11.2013	35	>35 m/6.11.2013	59	3-6 m9.11.2013
12	0-3 m/8.11.2013	36	3-6 m/6.11.2013	60	3-6 m/9.11.2013
13	0-3 m/9.11.2013	37	12-20 m/6.11.2013	61	6-12 m/9.11.2013
14	0-3 m/9.11.2013	38	12-20 m/6.11.2013	62	20-35 m/9.11.2013
15	6-12 m/9.11.2013	39	0-3 m/13.11.2013	63	>35 m/9.11.2013
16	6-12 m/9.11.2013	40	0-3 m/13.11.2013	64	>35 m/9.11.2013
17	6-12 m/9.11.2013	41	12-20 m/13.11.2013	65	0-3 m/12.11.2013
18	20-35 m/9.11.2013	42	12-20 m/13.11.2013	66	3-6 m/12.11.2013
19	>35 m/13.11.2013	43	12-20 m/13.11.2013	67	6-12 m/12.11.2013
20	>35 m/13.11.2013	44	12-20 m/13.11.2013	68	12-20 m/12.11.2013
21	>35 m/13.11.2013	45	20-35 m/13.11.2013	69	12-20 m/12.11.2013
22	12-20 m/9.11.2013	46	>35 m/13.11.2013	70	20-35 m/12.11.2013
23	12-20 m/13.11.2013	47	>35 m/13.11.2013	71	>35 m/12.11.2013
24	12-20m/13.11.2013	48	>35 m/13.11.2013	72	>35 m/12.11.2013

LAKE OHRID ADDITIONAL SAMPLING DATA (MACEDOIAN PART)

SUB BASIN	SAMPLING DATE	AIR TEMPERATURE (°C)	WATER TEMEPERATURE (°C)	SECHI DEPTH (M)	pH	OXYGEN (mg·l^{-1})	CONDUCTIVITY (µS)	WEATHER CONDITIONS	MOON
SB1 VELI DAB	19.10.2013	9.5	16.7	14.20	8.35	9.4	191.8	Clear, calm	Full moon
	04.11.2013	13.5	16.1	14.70	8.30	9.7	195.2	Clear, calm	No moon
SB2 ANDON DUKOV	08.11.2013	11.5	15.9	16.40	8.75	10.3	220	Clear, calm	No moon
	11.11.2013	11.1	15.8	16.00	8.55	10.1	224	Strong winds, waves	No moon
SB4 RADOZDA	05.11.2013	15.6	16.2	9.56	8.32	9.9	192.7	Overnight rain and storm; wind 22 m/s	No moon, cloudy
	06.11.2013	19.9	16.1	9.80	8.50	10.3	197	Overnight waves, lifting time calm	No moon, cloudy
SB3 CENTRAL PLATE	12.11.2013	Due to the storm no possibility for measuring	Due to the storm no possibility for measuring	Due to the storm no possibility for measuring	Due to the storm no possibility for measuring	Due to the storm no possibility for measuring	Due to the storm no possibility for measuring	Storm, strong winds, waves	cloudy
Pelagic net testing **VELI DAB**	19.10.2013	9.5	16.7	14.20	8.35	9.4	191.8	Clear, calm	Full moon clear

LAKE OHRID ADDITIONAL SAMPLING DATA (ALBANIAN PART)

SUB BASIN	SAMPLING DATE	AIR TEMPERATURE (°C)	WATER TEMEPERATURE (°C)	SECHI DEPTH (M)	pH	OXYGEN ($mg \cdot l^{-1}$)	WEATHER CONDITIONS	MOON
SB 5 LIN	06.11.2013	19.5	16.2	16.20	8.4	9.8	Overnight waves, lifting time calm	No moon, cloudy
	08.11.2013	13.5	16.1	15.70	8.50	9.7	Clear, calm	No moon
	09.11.2013	13	16	16.10	8.65	10.2	Clear, calm	No moon
	13.11.2013	11	15.7	15.80	8.55	9,8	Strong winds, waves	No moon
SB 6 HUDENISHT	05.11.2013	17,5	16,1	15,80	8,4	9,9	Overnight rain	No moon, cloudy
	06.11.2013	19	16,1	15,70	8,3	9,6	Overnight waves	No moon, cloudy
	13.11.2013	11,5	15,8	14,30	8,5	10,1	Clear, calm	No moon
SB 7 TUSHEMISHT	08.11.2013	13	16,2	11,30	8,4	9,6	Clear, calm	No moon
	09.11.2013	13	16,1	11,50	8,5	9,5	Clear, calm	No moon
	12.11.2013	No possibility for measuring due to the storm	No possibility for measuring due to the storm	No possibility for measuring due to the storm	No possibility for measuring due to the storm	No possibility for measuring due to the storm	Storm, strong winds, waves	cloudy

-FINAL REPORT-

PHYSICO-CHEMICAL INVESTIGATIONS OF THE WATER OF LAKE PRESPA AND RIVER GOLEMA

Transboundary Monitoring Program

Prepared by Dr Elizabeta Veljanoska-Sarafiloska
Hydrobiological Institute, Ohrid, R. Macedonia

June 2014

Contents

List of abbreviations

BOD_5 - biochemical oxygen demand
NO_2-N - nitrite-nitrogen
NO_3 - nitrate nitrogen
NH_3-N - ammonia-nitrogen
$TN_{Kjeldahl}$ - total nitrogen by Kjeldahl
TN - Total nitrogen
TP - total phosphorus
TSI - trophic state index
OCP - organochlorine pesticides

1. Introduction

The ecological system of lakes is highly dependent on the environment, i.e. on the physical, chemical and biological conditions in the watershed area. Human impact and the changes caused by human activities as well as natural changes could cause an increasing flow of nutrients. Nutrients especially phosphorus as limiting element of eutrophication can affect of trophic state of the water bodies. Ohrid and Prespa Lake belong to the largest and most important natural water ecosystems in the Balkan region. They belong to the Desaretian group of lakes and with their natural rarities and endemic species.

The classification of the water bodies is made according the Carlson index for trophic state (1977), OECD classification (1982) and Directive for Classification of Waters (1999)

The application of the Carlson's method for determination of the trophic state of water considers the following parameters:

- **Concentration of total phosphorus**
- **Secchi transparency**

2. Parameter and sampling points

Water samples were collected from following sampling sites on the lake:

MK I WWTP Ezerani (N 41° 00'962; E 21° 01'643);

MK II NW Ezerani (N 41° 00'131; E 20° 56'683), the bottom is very fine sand and mollusc shells, sampling point is near to the reed beds;

MK III NE Ezerani (N 41° 00'015; E 20° 58'656), the bottom is very fine sand and sampling point is near to the reed beds;

MK IV Ezerani littoral (N 41° 00'197; E 20° 57'741), the bottom is very fine sand and sampling point is near to the reed beds;

MK V Oteshevo (N 40° 58'692; E 20° 55'016), the bottom is sand, mud and mollusc shells;

MK VI Pelagic zone (N 40° 57'949; E 20° 57'513), the bottom is soft, fine silt

Additionally water samples were collected from the following sampling sites at the River Golema:

Golema 1 River Golema middle course (N 41° 05'040; E 21° 01'324), the bottom is man-made stone bad;

Golema 2 River Golema Inlet (N 41° 00'997; E 21° 01'767), the bottom is mud.

Sediment samples were collected from following sites: MK V (Oteshevo) and MK IV (Ezerani littoral).

Figure 1: Sampling sites Lake Prespa pelagic zone, littoral zone and main tributaries

Following physico-chemical parameters have been analysed within the investigations of the CSBL project:

- **transparency**
- **temperature**
- **pH**
- **conductivity**
- **dissolved oxygen**
- **biochemical oxygen demand (BOD_5)**
- **organic matter (through permanganate consumption)**
- **oxygen saturation**
- **total phosphorus**
- **nitrite-nitrogen (NO_2-N)**
- **nitrate-nitrogen (NO_3-N)**
- **ammonia-nitrogen (NH_3-N)**
- **$TN_{Kjeldahl}$ (total nitrogen by Kjeldahl)**
- **TN total nitrogen**
- **suspended solids**

3. Description of investigations

The sampling of water at all measuring stations was carried out in four campaigns, which are belonged to four different seasons as recommended in the WFD: April, July, October and January.

Samples for physico-chemical parameters have been taken during four sampling campaigns:

- first campaign in spring period 2013, the weather in this campaign was sunny with a light south wind
- second campaign in the summer period 2013, the weather in this campaign was sunny
- third campaign in the autumn period 2013, the weather in this campaign was sunny
- fourth campaign in the winter period 2014, the weather in this campaign was snow, wind with cloudy

Collecting of samples from pelagic was done by boat belonging to the HBIO. Two days were spent for collection of samples in each campaign (one for pelagic and one for rivers and littoral zone).

Ruttner bottle of 2,25 l (Hydro-bios, Kiel, Germany) was used for collecting the water samples from the littoral zone. The water samples from the pelagic zone (at two depths down the water column: 1 and 15 m), were collected with Niskin bottle (5l). The water sample from the river was collected directly into bottles. All sampling procedures fulfilled the requirements of official standard methodologies.

Samples for general chemical analysis were collected in polyethylene bottles. Samples for oxygen and BOD_5 were collected in Winkler bottles of 110-140ml and add 0.5 mL $MnSO_4$ followed by 0.5 mL KJ immediately after the sample collected. Samples for total phosphorus were collected in 130 ml polyethylene bottles and were filled completely with sample after rinsing them twice with the water to be analysed.

Samples for nitrogen compound (nitrite, nitrate, ammonia), should be collected in polyethylene bottles (500 ml) and were analysed immediately within 24h after collection. The best technique in nitrogen analysis is to do any filtrations within 5-10 hours of taking samples (which are kept cool and dark) and then quickly deep-freeze filtrates to -20 °C in polyethylene bottles.

Sediment samples were collected at the two sampling points Otesevo littoral and Ezerani littoral. The sampling was conducted with a Van-Veen grab sampler with a volume of 440 cm^3 (Hydro-bios, Kiel, Germany).

4. Transport and storage of samples

Bottles with samples were disposed in hand refrigerators at low temperature. Samples were transported to laboratory by car. The containers with samples were saved of any outside influences. Samples were transported to laboratory immediately. After their arrival in the laboratory they were disposed in refrigerators at 4°C till the analysis. Samples for total phosphorus should be kept in a cool dark place (4 °C) and not warmed to room temperature until the analysis has been to be commenced.

Sediment samples were stored and transported in field – refrigerators. Before analysis samples were stored at 4 °C for a maximum of seven days.

5. Analysis

All analysis have been done by standard methods (APHA-AWWA-WPCF 1980).

Transparency was measured by Secchi disk (secchi disk is a 20 cm diameter). Temperature was measured by reversing thermometer (Welch, 1948), pH was measured by WTW, Multilab 540 and conductivity by WTW conductometer LF 197. Temperature of water, transparency by Secchi disk, pH and conductivity were measured on the field.

Alkalinity was determined by titrating with standard sulphuric acid (0.02N) at room temperature using phenolphthalein and methyl orange indicator).

The organic matter content in the water expressed as permanganate consumption was determined by digestion in acid and by titration (Bether,1953).

Winkler method was used for analysis of dissolved oxygen and BOD_5. The fixation of dissolved oxygen was immediately after the sample collected in a winkler bottle with 0.5 mL $MnSO_4$ followed by 0.5 mL KJ.

Standard limnological methods (Beter, 1953; Ruttner, 1975; Golterman et al., 1978; Wetzel & Likens, 2000; APHA-AWWA-WPCF, 2005) were used for all titrimetric analyses.

The Total Organic Carbon (TOC) (index of the total amount of organic substances in water) was measured using a Total Organic Carbon Analyzer TOC – TOC ID.

Method 2540D in *Standard Methods for the Examination of Waters and Wastewaters,* 20th Edition (American Public Health Association, 1998) was used for total suspended solids. Therefore, a glass-fiber filter retains the suspended solids. After filtering the sample, the filter is dried to a constant weight at 103–105 °C.

Nitrates are quantitatively reduced from the water to nitrites by a cadmium-cooper couple (Stricland & Parsons, 1972; APHA-AWWA-WPCF 1980).

The procedure for determination of ammonia is performed by binding of the ammonia with hypohlorite in monochromine which in reaction with phenol gives p-aminophenol. In reaction with natrium nitroprusside it forms blue-coloured compounds (Solorzano 1969., Wetzel and Likens 1979). Water for ammonia nitrogen determination should be filtered.

Total nitrogen is the sum of organic nitrogen and inorganic nitrogen. In principle organic nitrogen but not nitrate or nitrite is reduced to ammonium during Kjeldahl digestion. The determination of ammonium in Kjeldahl digestion measures organic nitrogen + ammonium.

TN= organic nitrogen + ammonium + nitrite + nitrate

Kjeldahl digestion method (Strickland, J. D. H., Parsons, T.R., 1968; APHA-AWWA-WPCF, 1980) was used for determination of total nitrogen. The results are read on using Perkin-Elmer UV-VIS, spectrophotometer, wavelength 530 nm (nitrite and nitrate) and 640 nm (ammonia and total nitrogen by Kjeldahl).

Total phosphorus was analyzed by acid digestion of persulphate (at 121 °C, pressure 1 at, and time 1 hour) all forms of phosphates are altered to orthophosphate (Strickland and Parsons 1968; Menzel & Corwing, 1965; ISO/DIS 6878/1 : 1984).

The analysis for orthophosphate has been performed using the filtered water without acid digestion (mineralization before the chemical analysis). 130 ml polyethylene bottles should be filled completely with sample after rinsing them twice with the water to be analysed. The analysis should be commenced as soon as possible, preferably within 1/2 hr, certainly before 2 hr. (Strickland and Parsons 1968; ISO/DIS 6878/1 : 1984).

The results for total phosphorus and orthophosphate is read on Spectrophotometer Specord; model S-10, Carl Zeiss Jena, 880 nm.

The water for some parameters (nitrogen compounds, orthophosphate) was filtered through the glass filter 0,45μm, as soon as they come in laboratory.

The determination of organochlorine pesticides in the sediment was conducted using the modified EPA 8081A method. 70 to 80 g of fresh and well-homogenized sediment were placed in a glass container and mixed with a 1:1 mixture of hexane and acetone (*V*/*V*) and of methanol. Solid-liquid extraction was performed on a magnetic stirrer for 2 hours at room temperature without any thermal treatment of the sample.

Quantitative analysis for organochlorine pesticides was conducted with a gas chromatograph (Varian, USA) Model 3800, with an ECD detector and nitrogen as the carrier gas. The column used for separation of OCP was VA-1701 (VA-123073-20) with length 30 m; I.D. 0.32 mm; film thickness 0.25 mm, and temperature limits from – 20 °C to 280 °C (300 °C).

Classification of the water has been done according to Carlson index for trophic state (1977), OECD classification (1982) and Directive for Classification of waters (1999)

The application of the Carlson's method for determination of the trophic state of water which refers to the following parameters: concentration of total phosphorus and Secchi transparency, provides the numeric value of the trophic state index. The classification of the obtained numerical values of the indexes is according trophic scale proposed by Aizaki (1981).

6. Evaluation

6.1. Lake Prespa pelagial, littoral and River Golema.

Transparency (s.c. Secchi depth)

Transparency of Lake Prespa water varied between 3 m during winter period and 5.8 m during spring.

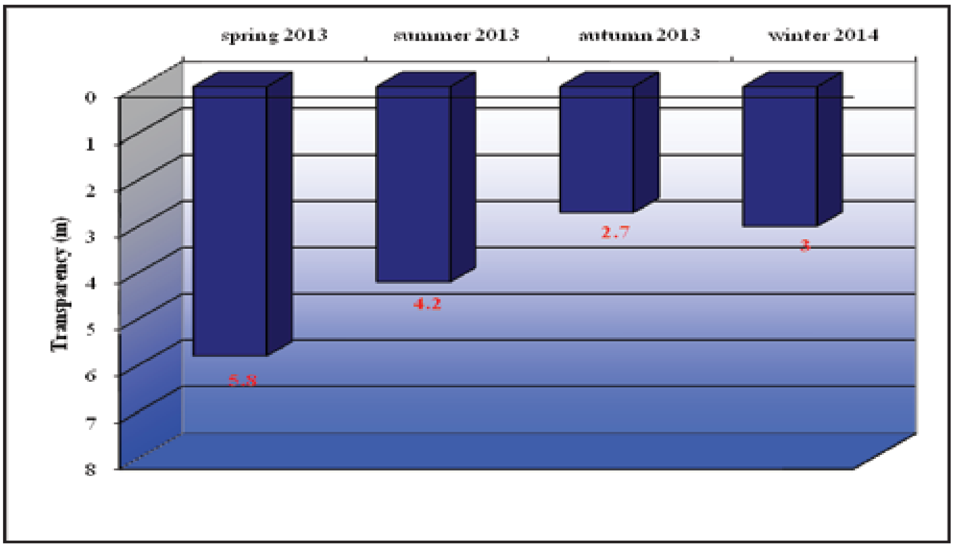

Figure 2: Transparency (Secchi depth) changes of Lake Prespa

Temperature

In general temperature increases during the heating period (spring and summer months). Highest values were measured during the summer period for all investigated points (25.6 °C Ezerani littoral). The lowest value for this parameter was measured in River Golema (0.34 °C, winter period). Water temperature in Lake Prespa typically varies from the surface to the deeper layers.

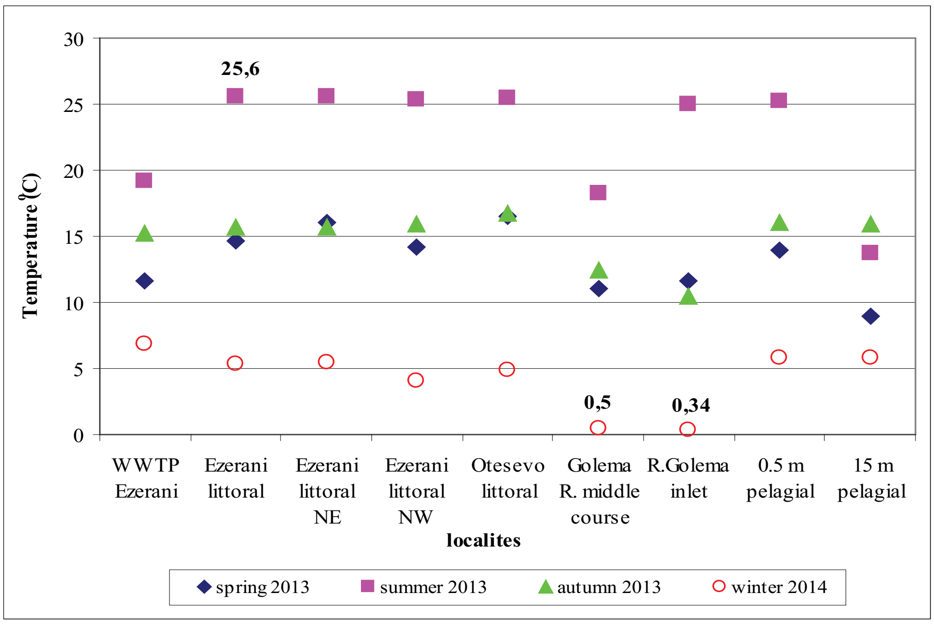

Figure 3: Temperature changes in the water from sampling points at Lake Prespa

pH

Minimal values for pH for all investigated period were registered in water samples collected from WWTP Ezerani (6.71 during spring) (Fig. 4).

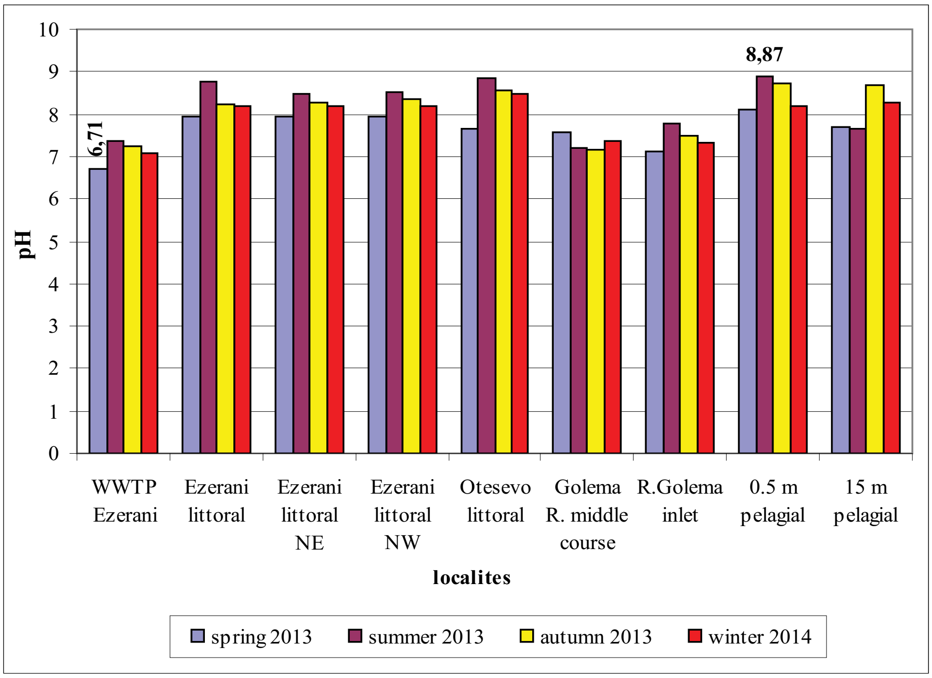

Figure 4: pH changes in the water from sampling points

Maximal values during all investigated period were registered at pelagic zone of Lake Prespa surface water. During summer period at this sampling point for pH was measured 8.87. (Fig. 4).

Conductivity

Values for conductivity are presented in figure 5. Conductivity in the water from sampling points varied between 607 mS cm^{-1} at WWTP Ezerani (summer period) and 143.2 mS cm^{-1} during winter.

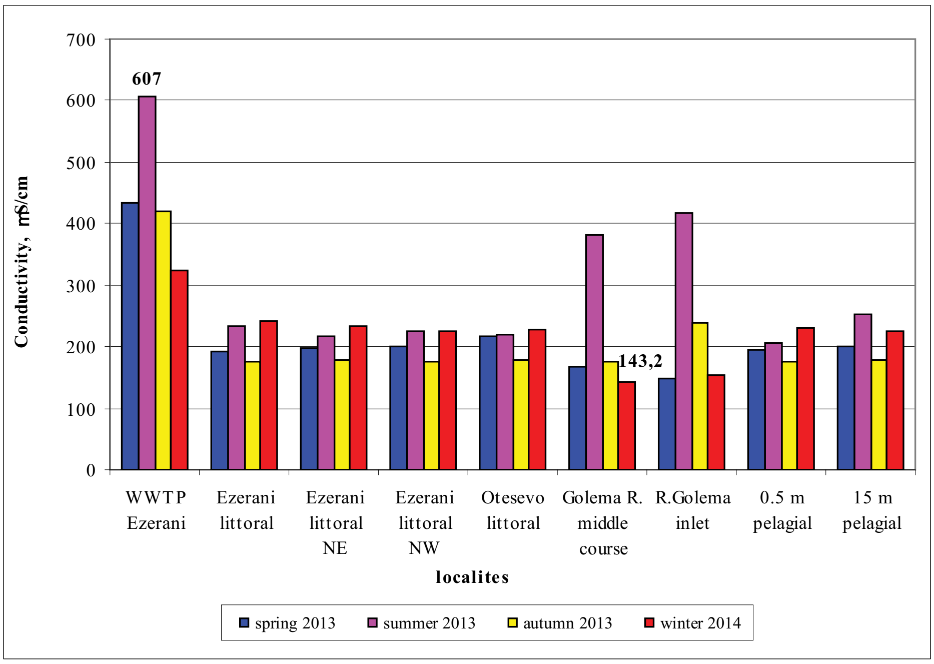

Figure 5: Conductivity changes in the water from sampling points

Total alkalinity

Total alkalinity values have varied from 239 mg l^{-1} $CaCO_3$ at WWTP Ezerani during the summer period to 82 mg l^{-1} $CaCO_3$ at River Golema middle course (during spring and winter) and River Golema inlet during winter period. Values for this parameter at water sample from the littoral zone of Lake Prespa during the investigated period are very closly.

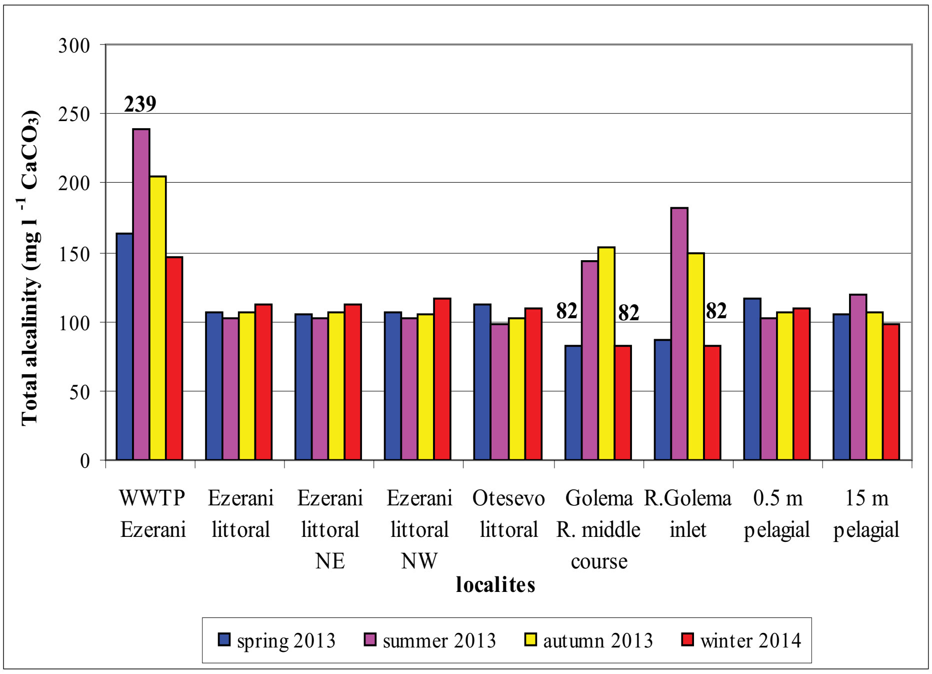

Figure 6: Total alkalinity changes in the water from sampling points

Total suspended solids

Minimal values for total suspended solids during the investigated period were registered at water samples collected from littoral zone of the Lake (0.012 g l^{-1} at Ezerani littoral, 0.013 g l^{-1} at Ezerani littoral NW and 0.015 g l^{-1} at Ezerani littoral NE and 0.017 g l^{-1} at Otesevo littoral during the winter).

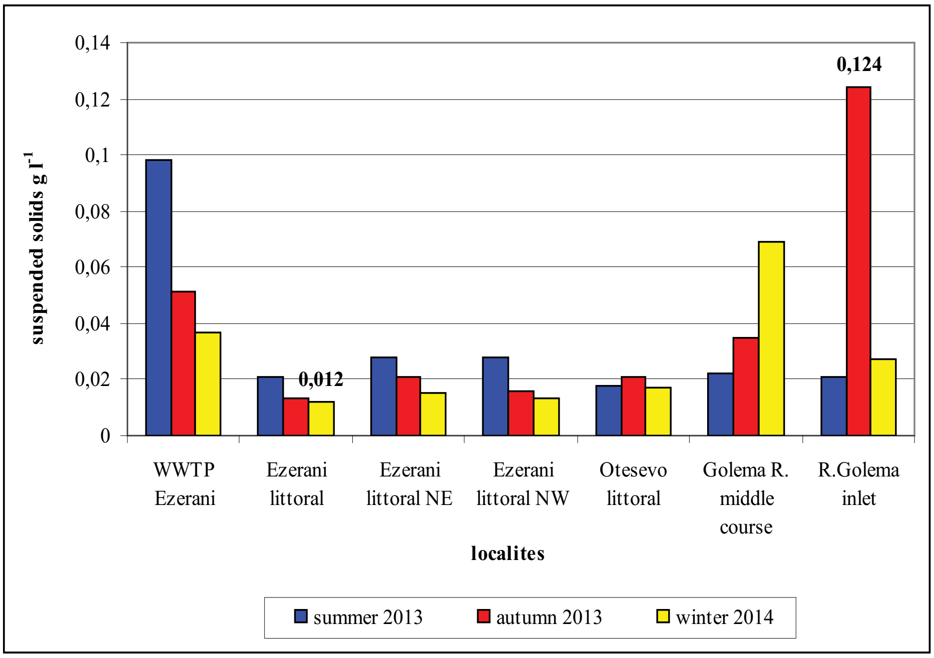

Figure 7: Suspended solids in the water from sampling points.

Maximal values for total suspended solids were registered in water samples collected from River Golema middle course, River Golema inlet and WWTP Ezerani. During the autumn maximal value for this parameter was 0.124 g l^{-1} (River Golema inlet). In summer period was registered 0.098 g l^{-1} for this parameter at WWTP Ezerani .

Dissolved oxygen

Oxygen is essential to the production and support of all life in the lakes i.e in all water ecosystems.

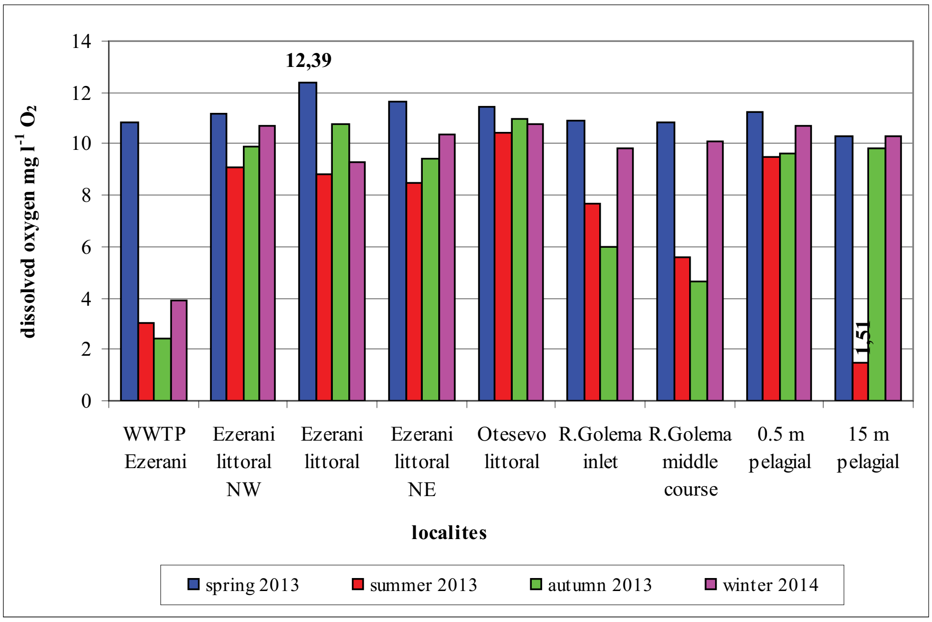

Figure 8: Concentration of dissolved oxygen in the water samples from sampling points

During all sampling campaigns it was observed that all depths have been supplied with oxygen sufficiently. All values for this parameter are above 8 mg l^{-1} O_2. Maximal value for this parameter was measured at water from Ezerani littoral, 12.39 mg l^{-1} O_2, during the spring period. Exception of this situation is observed in localities WWTP Ezerani during summer, autumn and winter period, River Golema inlet and middle course during summer and autumn and pelagial zone at 15 m depth (1.51 mg l^{-1} O_2) during the summer period, when were measured the lowest values for this parameter.

Oxygen saturation

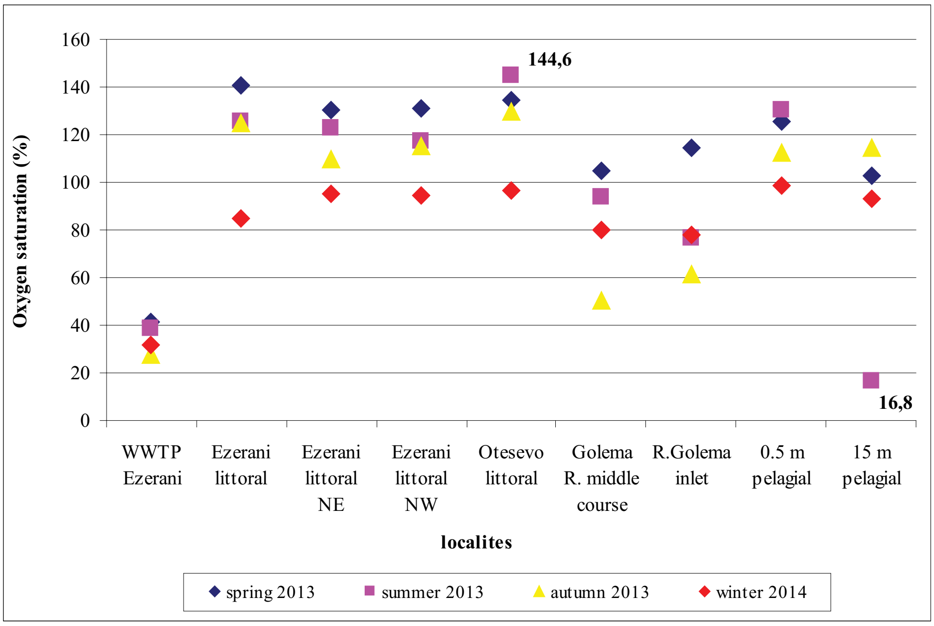

Figure 9: Oxygen saturation in the water from sampling points

Values for oxygen saturation in the water samples from littoral and pelagial zone of Lake Prespa and River Golema varied between 16.8 % at 15 m depth, pelagial zone and 144.6 % for Otesevo littoral during summer.

Biochemical oxygen demand (BOD_5)

BOD_5 is the amount of dissolved oxygen needed by aerobic biological organisms to break down organic material present in a given water sample at certain temperature over a specific time period. The term also refers to a chemical procedure for the determination of this amount. This is not a precise quantitative test although it is widely used as an indicator of the organic quality of water.The BOD value is most commonly expressed in milligrams of oxygen consumed per litre of sample during 5 days of incubation at 20 °C and is often used as a robust surrogate of the degree of organic pollution of water.

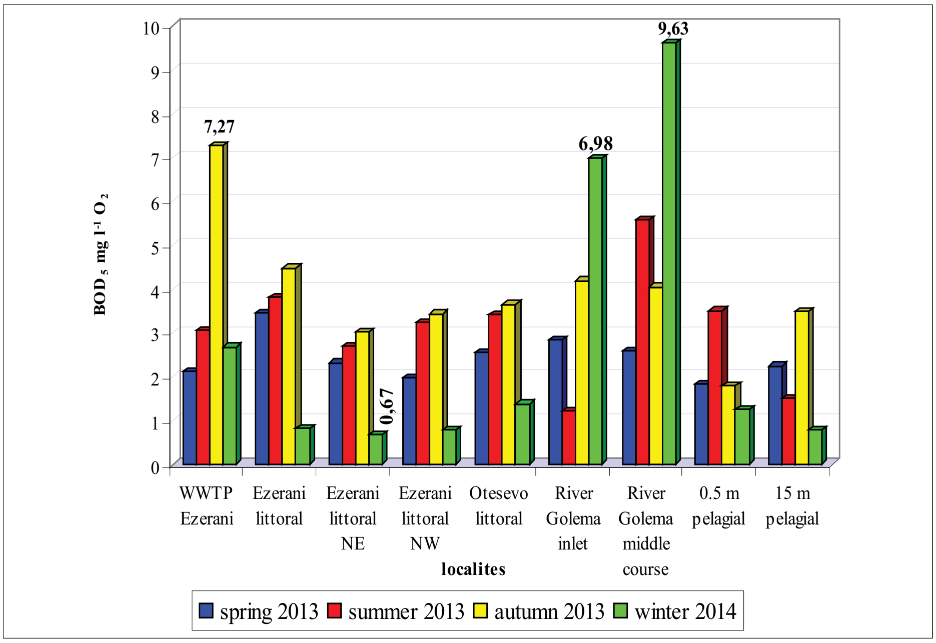

Figure 10: Biochemical oxygen demand in water from sampling point

Maximum values for BOD as indicator for organic loads were measured during summer and autumn in all sampling points from littoral and pelagial zone. Maximum value for BOD at water samples from River Golema was measured during winter period (6.98 mg l^{-1} O_2 at River Golema inlet and 9.63 mg l^{-1} O_2 at river Golema middle course). Highest value for this parameter has been registered in the water from the littoral Ezerani presumably as a reflection of the influence from the WWTP.

The lowest values for biochemical oxygen demand (BOD_5) during winter period were measured for Ezerani littoral (0.81 mg l^{-1} O_2), Ezerani littoral NE (0.67 mg l^{-1} O_2) and Ezerani littoral NW (0.79 mg l^{-1} O_2).

Organic matter

The consumption of $KMnO_4$ is an indirect measure for the quantity of organic biodegradable matter in the water (the quantity of consumed permanganate depends on the quantity of organic substance in the water and their chemical structure).

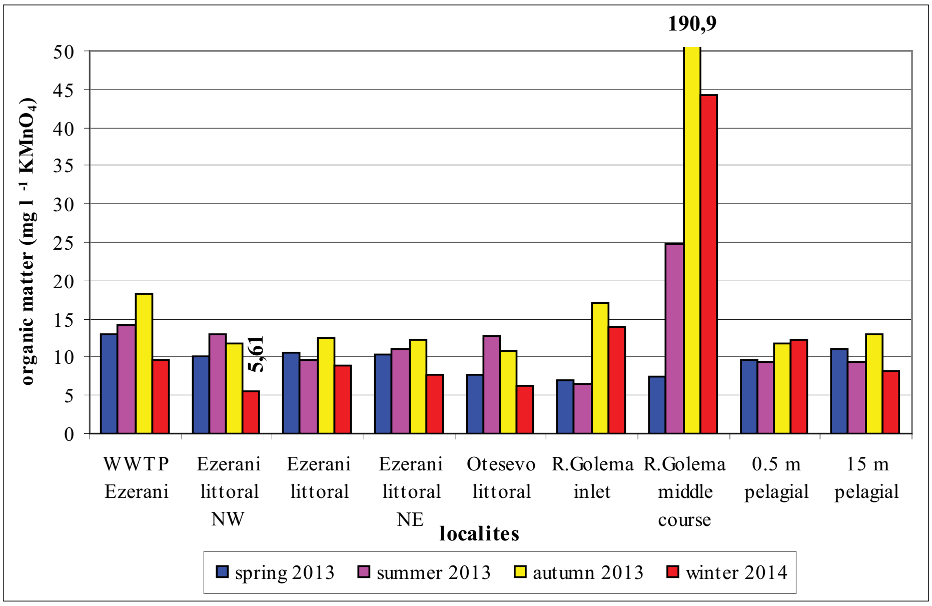

Figure 11: Organic biodegradable matter in water from sampling point

The obtained results for this parameter indicate that the highest values have been observed in River Golema middle course and River Golema inlet. In autumn, campaigns registered extremely high values for this parameter (190.26 mg l^{-1}) in River Golema middle course. This situation is due to the fact that these rivers pass through agricultural areas and settlements and they are "the end-recipients" of the industrial waste water, drainage water and sewage from households which are not connected with sewer and discharge directly into the river. Minimum value for this parameter was measured at water sample from Ezerani littoral NW (5.61 mg l^{-1}) during the winter period. During the all investigated campaigns the values for this parameter from the other sampling sites are below the 10 mg l^{-1}.

There is good correlation between the values obtained for BOD and organic matter in water samples (oxygen is used for oxidation the organic matter). So when high values are recorded for organic matter BOD values are also high. For example, maximal value for BOD for the winter period in River Golema middle course was 9.63 mg l^{-1}. In the same period, the maximal value for the content of organic matter was 44.18 mg l^{-1}.

Nitrogen compounds

The nutrient loading of the water from investigated points has been determined based on the concentration of two most important biogenic elements: total phosphorus and total nitrogen.

Nitrogen is beside phosphorus the relevant nutrient for the eutrophication of aquatic ecosystems. Tributaries are the biggest sources of nitrogen loads to Lake Prespa. In contrast to phosphorus nitrogen could also be an impact from the atmosphere.

There were analyzed several nitrogen compounds present in the water samples as: NH_3-N, NO_2-N, NO_3-N, $TN_{Kjeldahl}$, and total nitrogen. These nitrogen compounds take part in different physiological processes they directly depend of the processes of production and mineralization which parallel exist in the water.

Nitrite-nitrogen

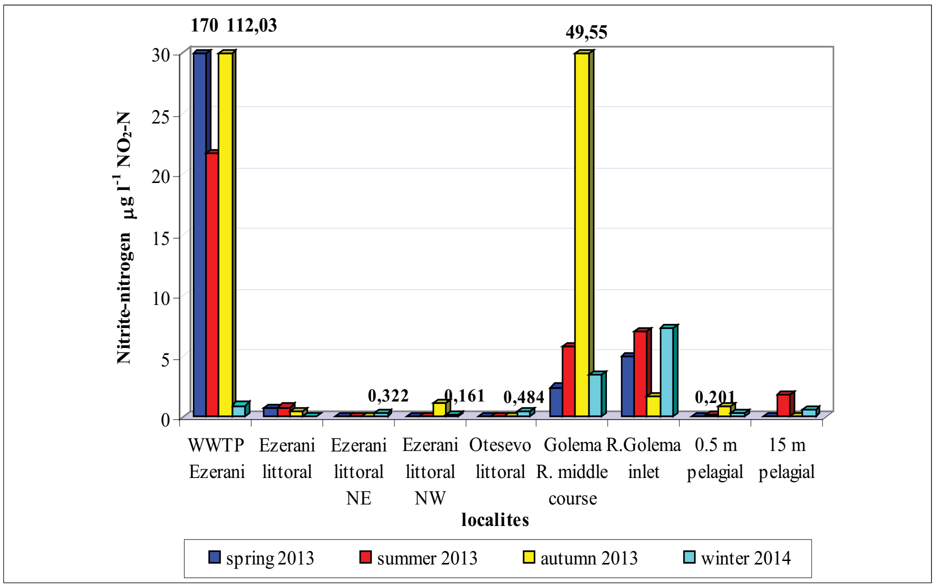

Figure 12: Nitrite - nitrogen concentration in water from sampling points

Concentration for this parameter were very low at all sampling campaigns (0.161 mg l-1 NO_2-N at Ezerani littoral NW, 0.322 mg l-1 NO_2-N at Ezerani littoral NE and 0.484 mg l-1 NO_2-N at Otesevo littoral during the winter, 0.201 mg l-1 NO_2-N at 0.5 m depth during the summer period). Maximal values were determined in water samples from WWTP Ezerani (170 mg l-1 NO_2-N in spring , River Golema middle course 49.55 mg l-1 NO_2-N in autumn and River Golema inlet 7.373 mg l-1 NO_2-N during the winter.

Nitrate-nitrogen

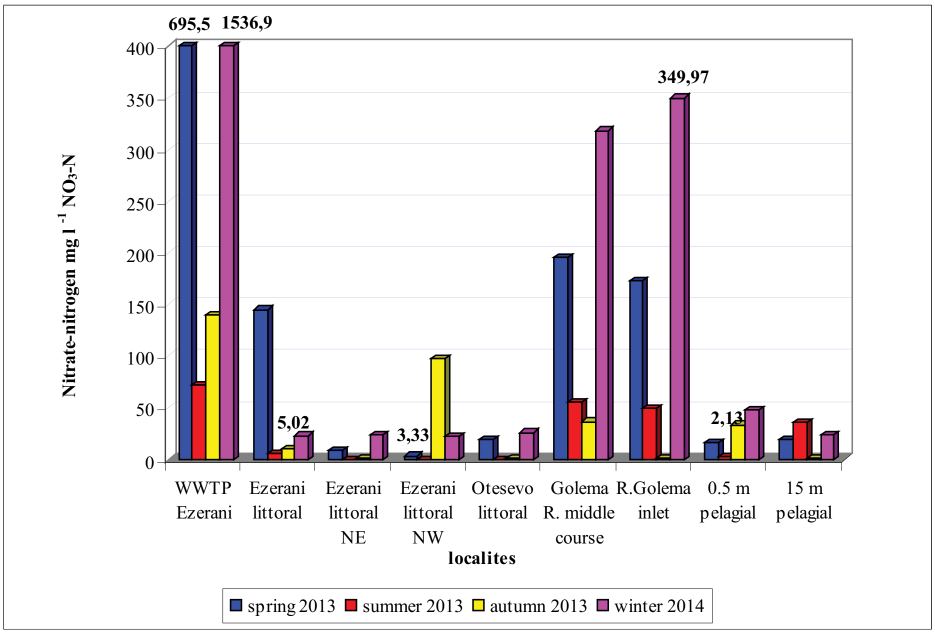

Figure 13: Nitrate - nitrogen concentration in water from sampling points

Concentrations of nitrate-nitrogen in the sampling points varied from 2.13 mg l-1 NO_3-N at 0.5 m depth during the summer period to 349.97 at River Golema inlet during the winter. Highest values for concentration of nitrate-nitrogen are registered in WWTP Ezerani and River Golema middle course and River Golema inlet. For WWTP Ezerani maximal value was 1536.9 mg l-1 NO_3-N during winter period, for River Golema inlet was 349.97 mg l-1 NO_3-N during winter and 318.16 mg l-1 NO_3-N was the maximum for River Golema middle course during the winter (Fig. 13).

Ammonia-nitrogen

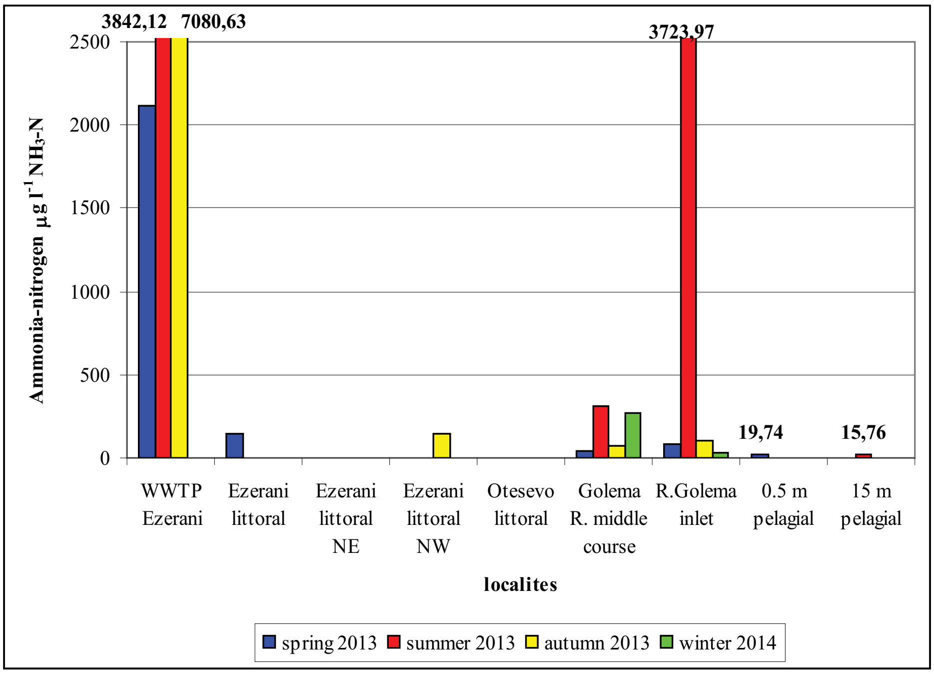

Figure 14: Ammonia - nitrogen concentration in water from sampling points

Maximal concentration for ammonia-nitrogen was measured at the outlet from WWTP Ezerani and River Golema middle sourse and inlet. For WWTP Ezerani maximal value was measured during autumn, 7080.63 mg l^{-1} NH_3-N, for River Golema inlet maximal value was measured during summer period, 3723.97mg l^{-1} NH_3-N and maximal value for River Golema middle course was 309.35 mg l^{-1} NH_3-N, during summer. Lowest value was registered at 15m depth, 15.76 mg l^{-1} NH_3-N.

Total Nitrogen by Kjeldahl

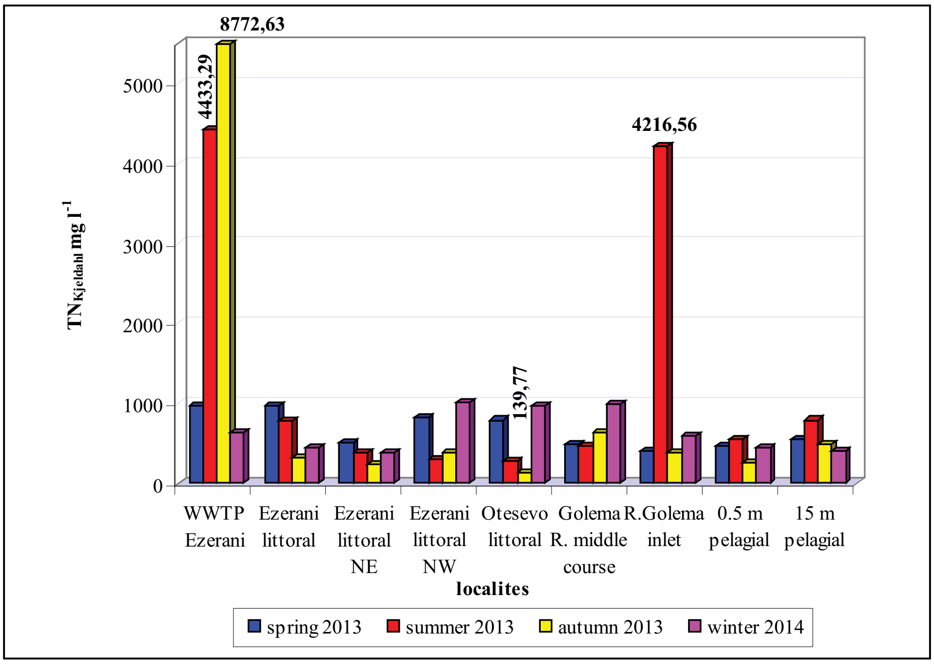

Figure 15: Total nitrogen by Kjeldahl concentration in water from sampling points

Concentrations of this parameter in the sampling points are very variable. The highest value was observed at the outlet of WWTP Ezerani (4433.29 mg l^{-1} $TN_{Kjeldahl}$, during autumn) and River Golema inlet (4216.56 mg l^{-1} $TN_{Kjeldahl}$, during summer). Lowest values for total nitrogen by Kjeldahl were measured at Ezerani littoral (NE and NW), Otesevo littoral and in the pelagial zone. During autumn at Otesevo littoral the minimal value was observed (139.77mg l^{-1} $TN_{Kjeldahl}$).

Total nitrogen

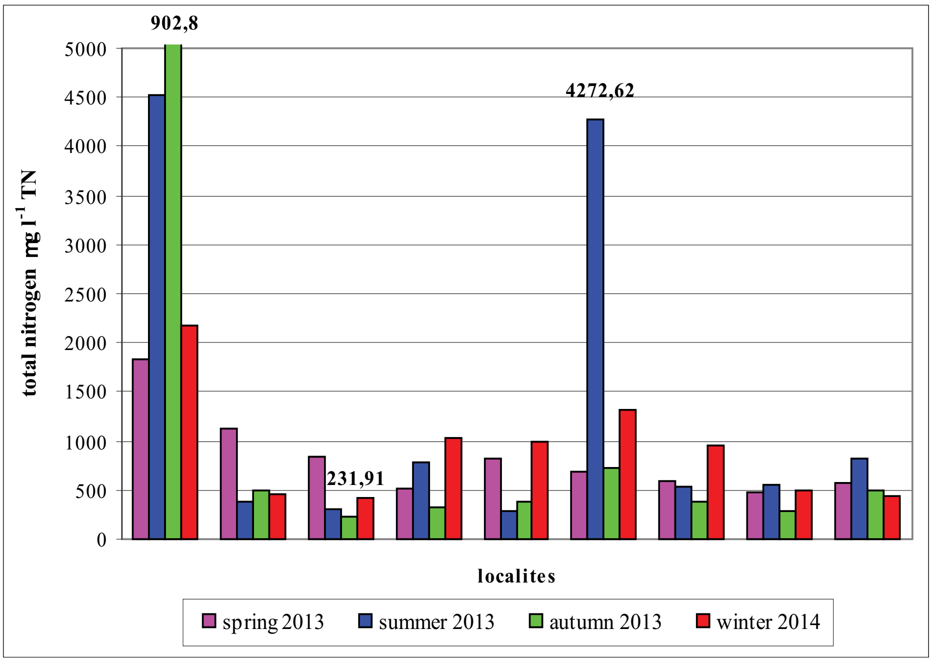

Figure 16: Concentration of total nitrogen in water from sampling points

Nutrient loads from River Golema middle course and in WWTP are very high. The maximum concentration of total nitrogen was observed in the outlet from WWTP (9024.38 mg l^{-1} TN) during summer. During the investigated period values for total nitrogen concentration in the littoral and pelagial zone of Lake Prespa were below 1000 (range of the concentration for this parameter is 231.91 mg l^{-1} TN for Ezerani NE to 1035.01 mg l^{-1} TN for Ezerani NW). Generally maximal values for all nitrogen were measured in River Golema middle course and WWTP.

According OECD classification and Directive for Classification of waters (Official Gazette No 18/99), based on concentration of total nitrogen, the water quality from the littoral and pelagia zone of Lake Prespa belong to II-IV class (meso-eutrophic state). River Golema is mainly IV class according Directive for Classification of waters (Official Gazette No 18/99) with meso-eutrophic character (OECD, 1982).

Total phosphorus

Total phosphorous as an essential nutrient It is the most limiting factor in eutrophication process of aquatic ecosystems. Even a small amount of phosphorus to a water body can have negative impact for the water quality. Those adverse effects include: algae blooms, accelerated plant growth and low dissolved oxygen from the decomposition of additional vegetation.

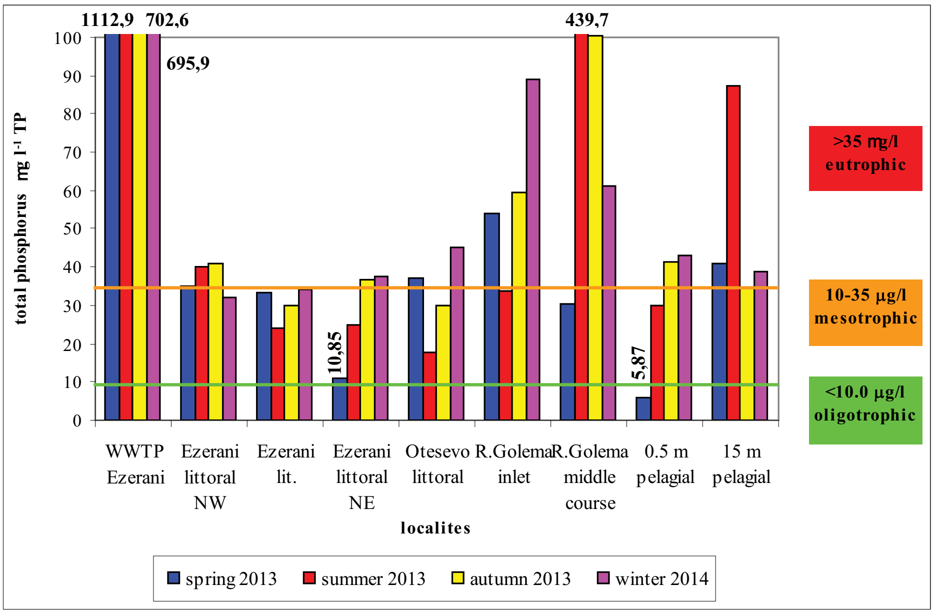

Figure 17: Concentration of total phosphorus in water from sampling points

Maximum values of total phosphorus were observed in the outlet of WWTP Ezerani (1112.9 mg l^{-1} TP during summer period), River Golema middle course (439.7 mg l^{-1} TP, summer period) and River Golema inlet (88.914 mg l^{-1} TP, winter). During the summer period the highest value for total phosphorus was registered in water from 15 m depth at the pelagial zone. **For phosphorus it is well known that anaerobic conditions above the sediment surface can strongly stimulate the release of phosphorus from the sediment. Due the fact that during the summer period at the 15 m depth od Lake (the bottom of the lake) concentration of dissolved oxygen is very low (there is a anoxic condition), the value for concentration of total phosphorus during this period is very high (87,203 mg l^{-1} TP).**

Generally many samples during the investigated period belong to mesotrophic state: Ezerani littoral, Ezerani littoral NW, Ezerani littoral NE, Otesevo littoral, pelagial zone 0.5 and 15 m, (according OECD classification of water), i.e. IV-V class according Directive for Classification of waters (Official Gazette No 18/99).

Trophic state index (TSI)

Carlson's trophic state index is summary, quick and indicative parameter for presentation of the trophic condition of certain water ecosystems. The calculation of this index is conducted by taking into consideration all characteristics (physical, chemical and biological) of the water represented via suitable researched parameters: Secchi depth (SD), total phosphorus concentration (TP) and chlorophyll *a* concentration (Chla).

Concentration of total phosphorous as a chemical indicator and the transparency as a physical parameter are used as indicators of the trophic condition of the Lake Prespa. According to the numeric values for the Carlson's Trophic State Index (TSI), based on secchi depth, during the investigation, Lake Prespa usually belong to **mesotrophic state**

According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus during the investigated period littoral zone of Lake Prespa belong to the **meso-eutrophic state**. According to the requirements of the Water Framework Directive we can define the status of Lake Prespa as moderate.

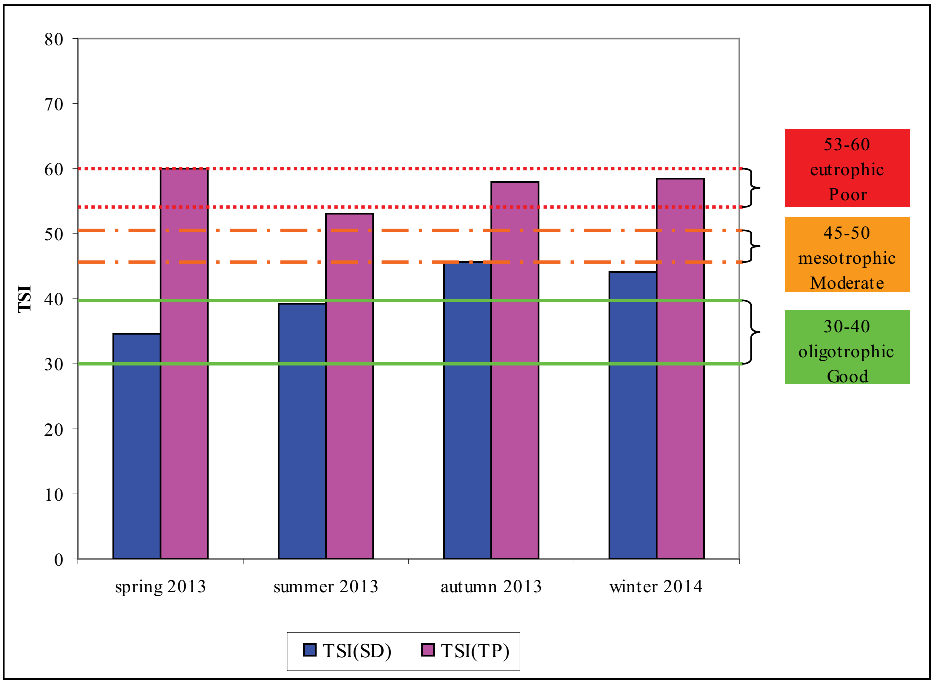

Figure 18: Trophic state index (TSI) for Lake Prespa pelagial zone

The values of the Trophic State Index (TSI) at the littoral zone of Lake Prespa based on concentration of total phosphorus indicate to a great seasonal variability .

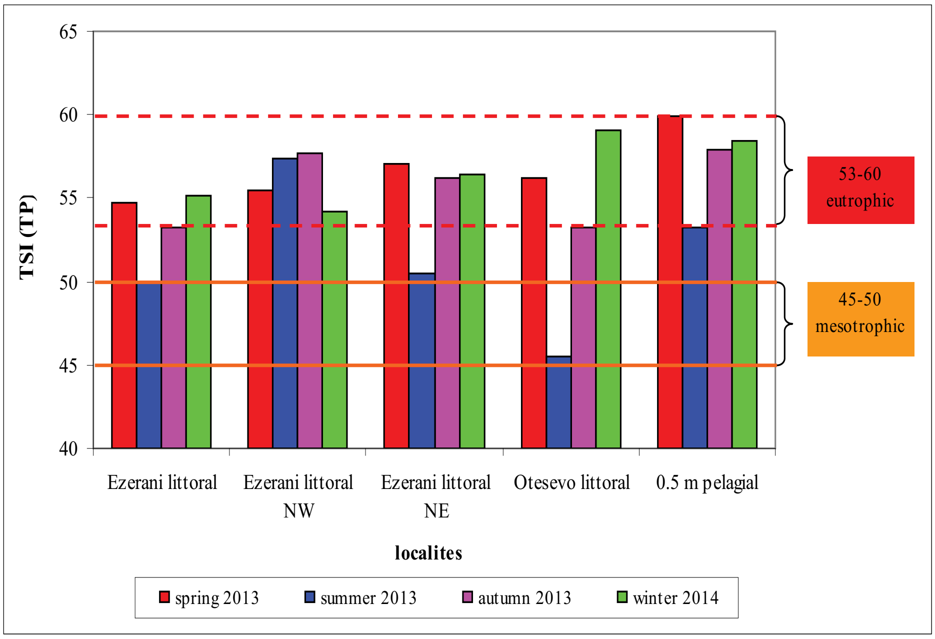

Figure 19: Trophic state index for Lake Prespa littoral zone

Sediment analysis

The organochlorine pesticides measured in sediment sample were: *gamma*-HCH (γ-HCH), ΣHCH (sum of α-isomer, β-isomer and δ-isomer, endosulfan (total of α and β endosulfan), DDT metabolites (*p,p'*–DDE, *p,p'*–DDD and *p,p'*-DDT). The obtained results show that the most abundant group of OCPs in analyzed sediment samples is total DDT (sum of p,p'DDT; p,p'DDD, p,p'DDE). The content of total DDT at sediment sample from Ezerani (3.01 mg kg^{-1} dry sediment) is higher than the content of this form estimated in sediment from Otesevo (1.824 mg kg^{-1} dry sediment). The mean value for the content of total detected organochlorine pesticides in sediment sample from Ezerani is 7.86 mg kg^{-1} dry sediment. For Otesevo mean value for the content of total detected organochlorine pesticides in sediment sample is 5.646 mg kg^{-1} dry sediment. The detected concentrations are clearly below toxic thresholds and consequently severe effects on the endemic species of Lake Prespa are not very likely.

Persistent organic pollutants (POPs), including organochlorine pesticides (OCPs), are of global concern because of their toxicity, resistance to degradation, potential for long-term transport and their tendency to accumulate in fatty tissues (lipophilicity), the latter of which renders them likely to bioaccumulate through food chain. The potential risks they pose to the environment and to human health are so serious that international treaties, e.g. the United Nations Aarhus Protocol (UN, 1998) and the Stockholm Convention (UNEP) aimed at elimination or restriction of their production and use have been established. In order to protect human health and environment from the influence of these pollutants, the Republic of Macedonia has also signed and implemented the Stockholm Convention in 2002.

6.2. Classification

- According to the OECD classification and Directive for Classification of waters (Official Gazette No 18/99), based on concentration of total nitrogen, the water quality from the littoral zone of Lake Prespa belongs to II-IV class (meso-eutrophic state). River Golema is mainly IV class, according to the Directive for Classification of waters (Official Gazette No 18/99) has it a meso-eutrophic character (OECD, 1982).
- Due the fact that during the summer period at the 15 m depth of the lake (the bottom of the lake) the concentration of dissolved oxygen is very low (there is a anoxic condition), total phosphorus concentration is extremely high (87,203 µg l-1 TP). The phosphorous concentrations at the other sampling points (Ezerani littoral, ezerani littoral NW, Ezerani littoral NE, Otesevo littoral, pelagial zone 0.5 and 15 m) shows mesotrophic status.
- According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus and the Secchi depth during the investigated period Lake Prespa is in mesotrophic state with tendency to eutrophic state. Lake Prespa based on all applied classifications (OECD fixed boundary system (1982) and Directive for classification of waters), is mainly in **mesotrophic state**. According to the Water Framework Directive Lake Prespa has a moderate status but **at risk** to fail the environmental objectives.
- According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus during the investigated period littoral zone of Lake Prespa belong to the **meso-eutrophic state.** According to the Directive for Classification of Waters (RM N° 18/99), mainly indicate to the water of III and IV category. According to OECD classification of water, values for TP belong to **meso-eutrophic** state and hyper-eutrophic state for water from WWTP and R. Golema middle course.

7. Recommendations

Considering that fifty years ago Lake Prespa has mostly oligotrophic conditions investigations within the CSBL project has demonstrated that Lake Prespa is still in an intensive process of eutrophication. Natural processes like the water level declination strengthen the process of deteriation of the trophic state. The changes in the volume of the Lake have a direct effect on the concentrations of dissolved nutrients. The intensive influence of the rivers which are flowing into Lake Prespa which is manifested in the lake littoral zone is a potential risk for the water status of the pelagic zone Furthermore dramatic changes have been observed in the littoral zone of the Lake Prespa especially near the confluence of River Golema. The future state monitoring programme has to consider the requirements of the WFD as well as of the Macedonian water law. It must involve the observation of these development as well as further investigations to assess the impact of rehabilitation measures which should start soon as possible.

8. Summary

The high nutrient concentrations in the Prespa tributaries reflect the surrounding intensive agriculture use and settlements in the watershed. Furthermore, insufficient waste water treatment and leakages in the sewer system have an heavy impact to the water status. Highest nutrient loads come from the discharge of WWTP Ezerani and river Golema (diffuse source from agriculture and domestic water).

According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus and the Secchi depth during the investigated period Lake Prespa is in mesotrophic state with tendency to eutrophic state. Lake Prespa based on all applied classifications (OECD fixed boundary system (1982) and Directive for classification of waters), is mainly in mesotrophic state.

According to the numeric values for the Carlson's Trophic State Index (TSI), calculated on basis of the total phosphorus during the investigated period littoral zone of Lake Prespa belong to the meso-eutrophic state. According to the Directive for Classification of Waters (RM No 18/99), mainly indicate to the water of III and IV category. According to OECD classification of water, values for TP belong to meso-eutrophic state and hyper-eutrophic state for water from WWTP and R. Golema middle course.

Persistent organic pollutants (POPs), including organochlorine pesticides (OCPs) are of global concern because of their toxicity, resistance to degradation, potential for long-term transport and their tendency to accumulate in fatty tissues. The detected concentrations for organochlorine pesticides at sediment samples are clearly below toxic thresholds and consequently severe effects on the endemic species of Lake Prespa are not very likely.

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Hydrobiological Institute Ohrid

Phytoplankton of Lake Prespa

–Final Report –

Dr. Suzana Patceva
Hydrobiological Institute - Ohrid

June, 2014

Contents

1. INTRODUCTION

Phytoplankton forms the base of many lake food webs. They consist of unicellular and colonial algae (from < 1 µm to >500 µm) floating in the water within the euphotic zone where light is available for photosynthesis.

Phytoplankton species represent the most sensitive biological components of water ecosystems that respond first to changes in nutrient concentrations. Regarding this fact, phytoplankton investigations are included in all monitoring projects of aquatic ecosystems.

The EU Water Framework Directive (WFD; 2000/60/EC) includes phytoplankton as one of four biological elements to be used in the assessment of the ecological status of surface waters.

Chlorophyll *a* is the green pigment in phytoplankton which allows photosynthesis to take place.

The Chlorophyll *a* concentration is an indicator of phytoplankton biomass, its concentration being proportional to the total biomass of phytoplankton. Additionally, the chlorophyll *a* concentration is one of the most indicative parameters of water trophic state.

2. METHODS

The phytoplankton analyses at Lake Prespa consisted of the following parameters:

- **Phytoplankton composition - Phytoplankton taxa present**
- **Phytoplankton abundance – Number of individuals per volume of water (cell/l)**
- **Diversity of phytoplankton community - Diversity index (Shannon-Wiener index)**
- **Bloom metric - Pielou's evenness index (shows how equal communities are numerically)**
- **Bloom metric - Cyanobacterial abundance**
- **Phytoplankton biomass – Chlorophyll *a* concentration(indirect assessment)**
- **Trophic State Index (TSI)**
- **Dominant species**

Sampling points

Investigations of phytoplankton and chlorophyll *a* concentrations in Macedonian part of Lake Prespa comprised samplings from 4 sampling points in the littoral zone and 2 sampling points in the pelagic zone.

The sampling points in Macedonia were:

LITTORAL ZONE (4)

- **WWTP Ezerani (Ezerani lit.)**
- **NW Ezerani**
- **NE Ezerani**
- **Oteshevo**

PELAGIC ZONE (2)

- **0.5 m**
- **15 m**

Sampling campaigns

Phytoplankton samples were taken during two sampling campaigns: the first one in spring (April 2013) and the second one in summer (July 2013).

The weather in spring was sunny with a light south wind and temperatures of surface water varied between 13.9 and 16.5 °C.

The weather in summer was sunny, water temperatures varying of the surface water in different sample points varied between 25.2 and 25.6°C.

Sampling and analyses

Sampling and analyses were done according to the joint CSBL – Protocol for phytoplankton. For qualitative and quantitative analyses of phytoplankton and chlorophyll *a* concentration, samples were taken with a Niskin water sampler. Samples for phytoplankton analyses were preserved immediately at the sampling site by adding 4% *formaldehyde*.

1. Qualitative (Identification of phytoplankton taxa) and quantitative analyses (enumeration of individuals per volume of water) were done according to Utermöhl (1958) using an inverted microscope LW101-2 trinocular, with epi-illumination module and camera, OmniVID, 8.0MP.

2. Chlorophyll *a* was analysed according to ISO 10260 (1992) using a spectrophotometer UV-VIS SPECORD 10 (Zeiss) after an extraction with 90% ethanol.

3. The calculation of the trophic state index (TSI) was done according to Carlson (1977) while evenness index (bloom metric) and the diversity index were calculated according to Pielou (1969) and Shannon-Wiener, respectively.

Figure 1. Sampling points for phytoplankton analyses in Lake Prespa

3. EXISTING DATA AND GAPS

Initial phytoplankton investigations at Lake Prespa began at the beginning of the XX century. In the second half of the last century phytoplankton investigations were conducted only occasionally (Kozarov, 1959, 1960, 1972; Mitic, 1996; Mitic et al., 1997). From 2001 to 2003 comparative analyses of phytoplankton communities and the trophic state of Lakes Ohrid and Prespa were conducted (Patceva, 2005). Water samples were collected from four depths in the pelagic zone of Lake Prespa with seasonal and monthly sampling periods (July- September). These studies included analyses of the qualitative and quantitative composition of phytoplankton and the chlorophyll *a* content and the correlation of these parameters with nutrient concentrations (phosphorus and nitrogen). They revealed significant changes in phytoplankton composition compared to previous studies owing to eutrophication (Patceva & Mitic, 2006).

In 2010 analyses of diatom assemblages in different core layers of Lake Prespa to reveal possible changes in dominant planktonic or benthic taxa and thus deduce the corresponding changes of environmental conditions forced by human activities and analyses of plankton communities during summer months were conducted (Prespa Lake Watershed Management Plan, GEF/UNDP National Prespa Park Project).

4. RESULTS

4.1. Phytoplankton composition

The phytoplankton species collected during the spring and summer campaigns belong to 6 divisions:

- **Cyanophyta**
- **Bacillariophyta**
- **Chlorophyta**
- **Chrysophyta**
- **Pyrrophyta**
- **Euglenophyta**

Most phytoplankton species identified belong to the Bacillariophyta and Cyanophyta investigations showed distinct seasonal differences in phytoplankton composition.

Bacillariophyta were the dominant group in spring, contributing between 93% and 98% of the total phytoplankton biomass in the pelagic zone and between 76% and 84% of the total phytoplankton biomass in the littoral zone. An exception was sampling point Ezerani NE where green algae (Chlorophyta) were the dominant group of algae in the spring period contributing 84% of the total phytoplankton biomass.

In summer Cyanophyta (blue-green algae) were the dominant algal group at all sampling points with the exception of samples from 15 m depth in the pelagic zone and Ezerani NE where Chlorophyta (green algae) dominated comprising 67% and 44% of the total number of individuals, respectively. The proportion of Cyanophyta was the lowest at 15m depth in the pelagic zone while it ranged between 21% (Ezerani NE) and 35% (Ezerani littoral) at the other sampling points.

The proportion of Pyrrophyta in summer was the highest at surface level (35%) and the lowest in Ezerani littoral (2%). The latter group of algae has the highest growth rates in summer at high temperatures and light intensity. The proportion of Chrysophyta was generally very low (<1%). This taxonomic group is characteristic of oligotrophic lakes.

The overall composition of phytoplankton communities at Lake Prespa is typical for lakes in the process of eutrophication.

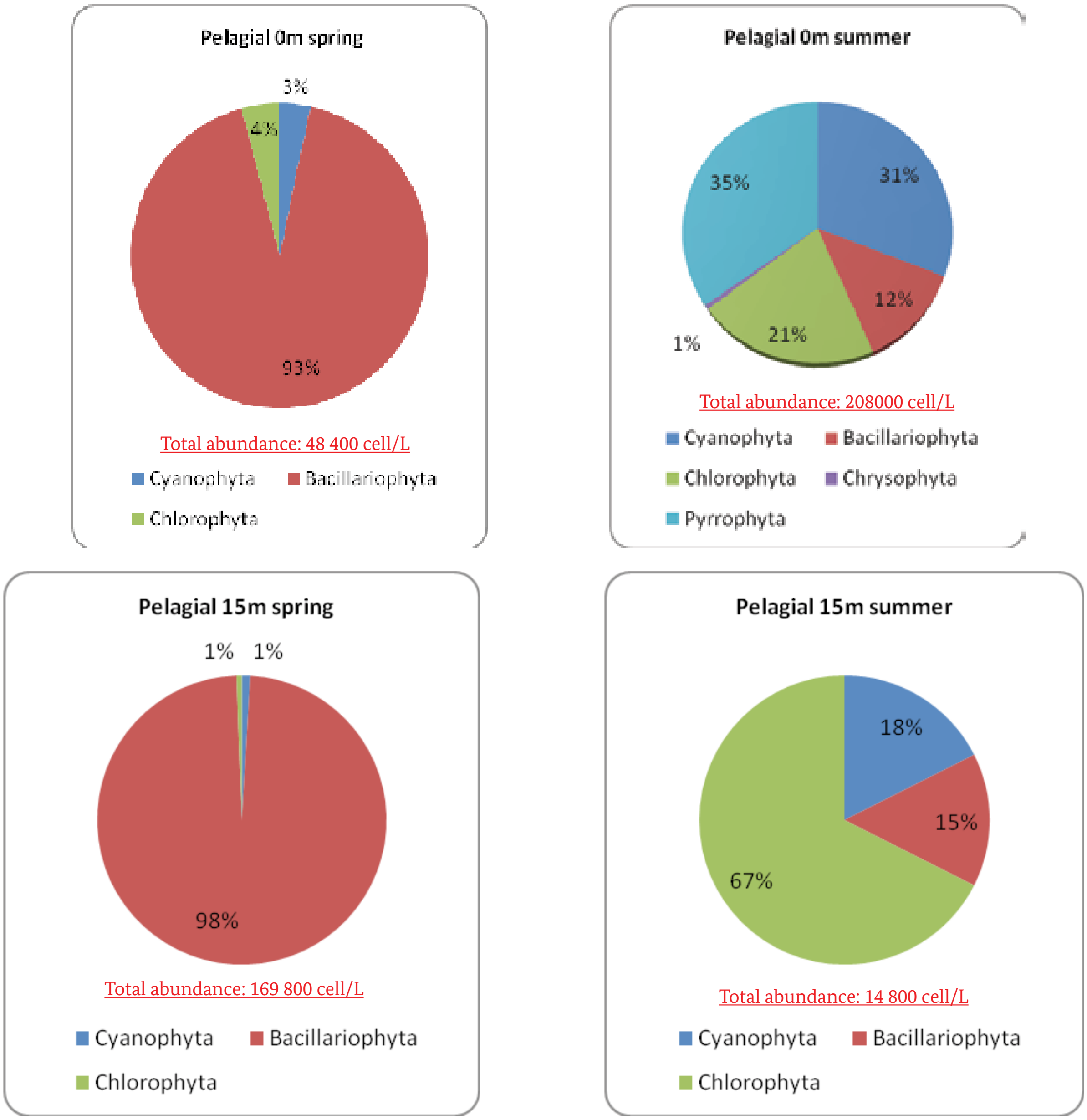


Figure 2. Percentage of algal divisions in the total number of identified species in the pelagic zone of Lake Prespa

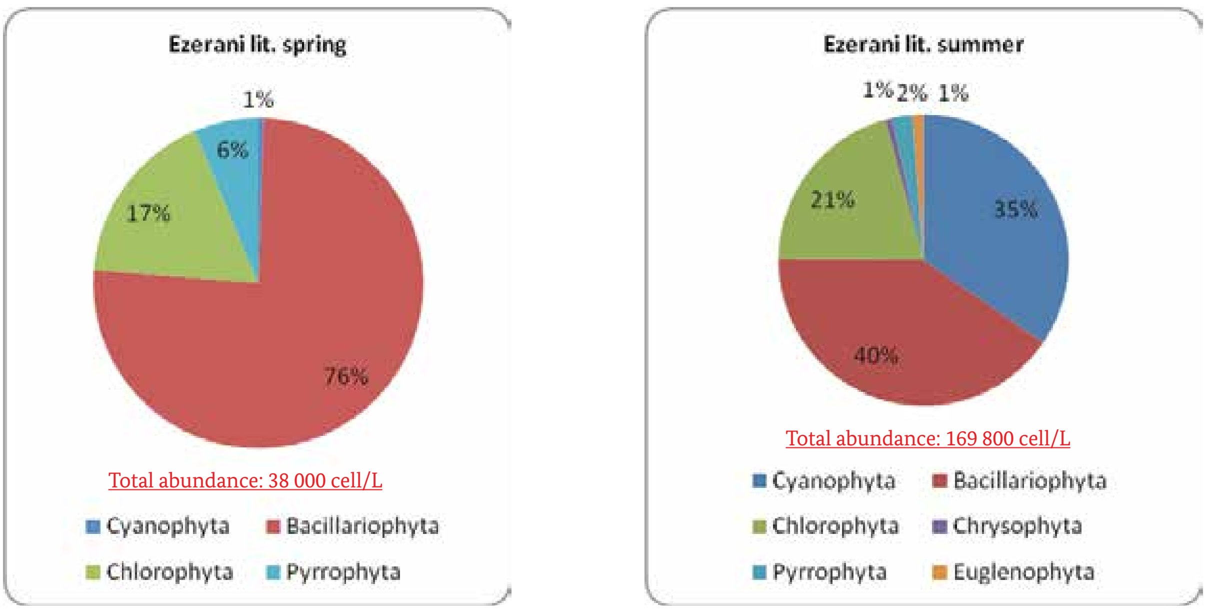


Figure 3. Percentage of algal divisions in the total number of identified species in WWTP Ezerani (Ezerani lit.)

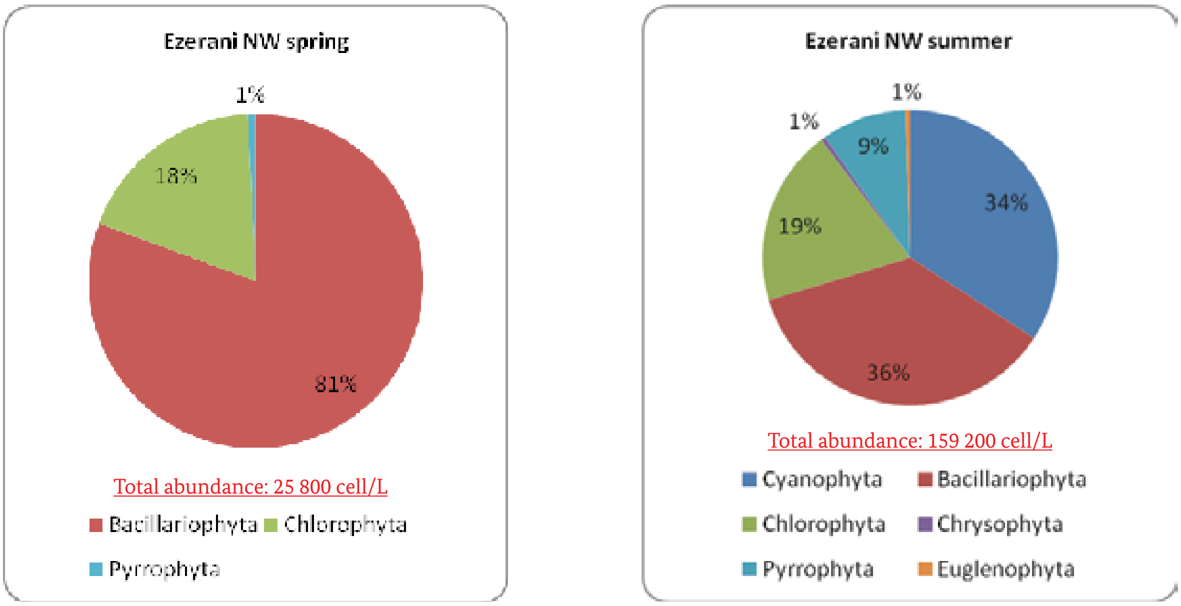


Figure 4. Percentage of algal divisions in the total number of identified species in Ezerani NW

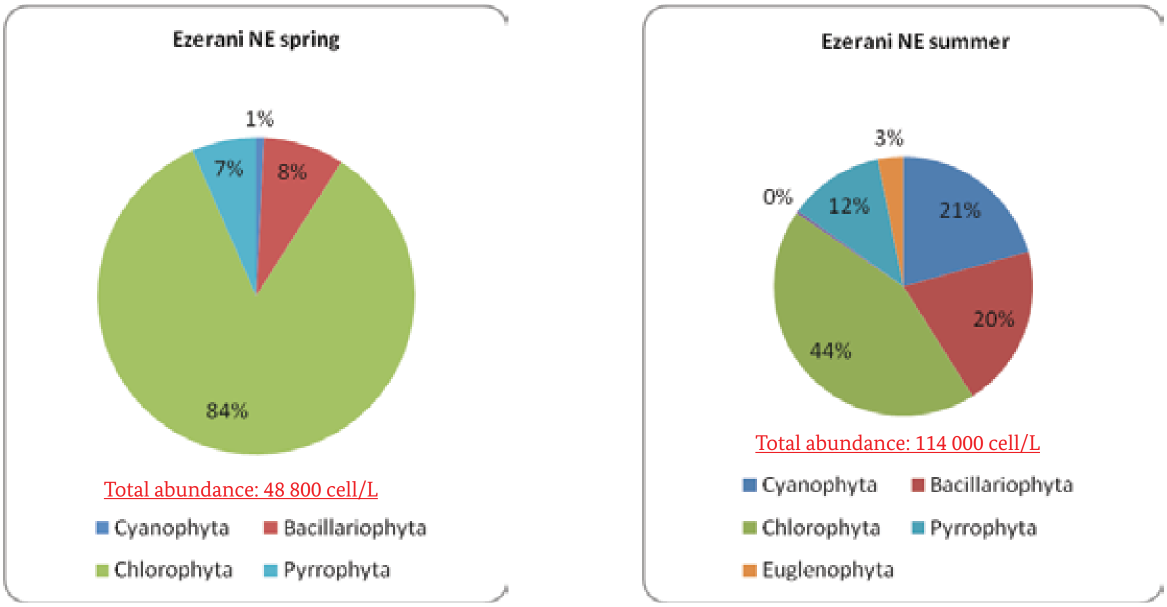


Figure 5. Percentage of algal divisions in the total number of identified species in Ezerani NE

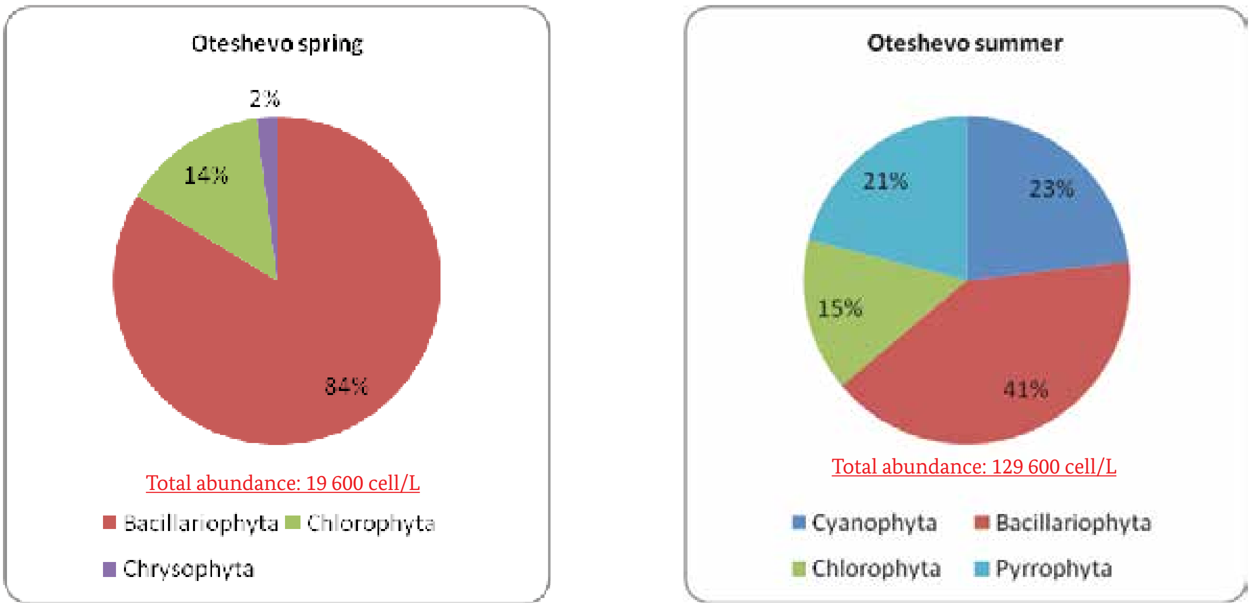


Figure 6. Percentage of algal divisions in the total number of identified species in Oteshevo

4.2. Phytoplankton indices

Diversity index is a quantitative measure that reflects how many different types (such as species) there are in dataset, and simultaneously takes into account how evenly the basic entities (such as individuals) are distributed among those types. The value of diversity index increases both when the number of types increases and when evenness increases. Theoretical maximum value for H is possible if all species in the sample were equally abundant. Evenness index refers to how close in numbers each species in an environment are.

In summer, the number of phytoplankton species was significantly higher than in spring at all sampling points. Likewise, **diversity** and **evenness index** were also higher in summer than in spring (Tab. 1, 2).

Table. 1. Species number, diversity index and evenness index at 6 sampling points in the Macedonian part of Lake Prespa during the first (spring) sampling campaign

	Pelagial 0m	Pelagial 15m	Ezerani lit.	Ezerani NW	Ezerani NE	Oteshevo
Species number	6	6	16	6	11	8
Diversity index (H)	0.41	0.11	1.36	0.79	0.98	0.86
Evenness index (J)	0.23	0.06	0.49	0.44	0.41	0.41

Table. 2. Species number, diversity index and evenness index at 6 sampling points in f the Macedonian part of Lake Prespa during the second (summer) sampling campaign

	Pelagial 0m	Pelagial 15m	Ezerani lit.	Ezerani NW	Ezerani NE	Oteshevo
Species number	17	7	36	25	26	16
Diversity index (H)	2.02	1.21	2.06	2.15	2.58	1.80
Evenness index (J)	0.71	0.62	0.58	0.67	0.79	0.65

The highest species number was recorded at Ezerani littoral (WWTP) in summer and the lowest in the pelagic zone and Ezerani NW in spring.

The highest diversity and evenness index (J) were found at Ezerani NE in summer and the lowest at 15 m in the pelagic zone in spring.

The lowest diversity and evenness index were observed in the pelagic zone at 15 m depth. The low values were due to a low number of species and the predominance of one species *Cyclotella ocellata* which accounted for 98% of the total phytoplankton abundance.

The highest diversity and evenness index were observed at Ezerani NE in July, which was due to a large number of species (26) and the absence of dominant species.

4.3. Phytoplankton abundance

The total abundance of phytoplankton was considerably higher in summer than in spring except for the pelagic zone at 15 m of depth where it was higher in spring, with *Cyclotella ocellata* as the dominant species (Fig. 7).

In summer, the highest total abundance of phytoplankton was found in the pelagic zone at 0 m of depth (Fig. 6). The dominant species were *Peridinium cunningtonii* (Pyrrophyta) and *Lyngbya limnetica* (Cyanophyta). At the same time, total abundance was lowest at 15 m depth.

In the littoral zone, the highest abundance was observed in summer at Ezerani littoral (WWTP) and Ezerani NW with domination of *Cyclotella ocellata* (Bacillariophyta) and *Lyngbya limnetica* (Cyanophyta).

Significant difference in dominant species was observed at locality Ezerani NE where was identified *Chlamidomonas* sp. (Chlorophyta) as dominant species in the first (spring) sampling campaign and *Chlamiydocapsa planctonica* (Chlorophyta) was observed at the second sampling campaign (Tab. 3).

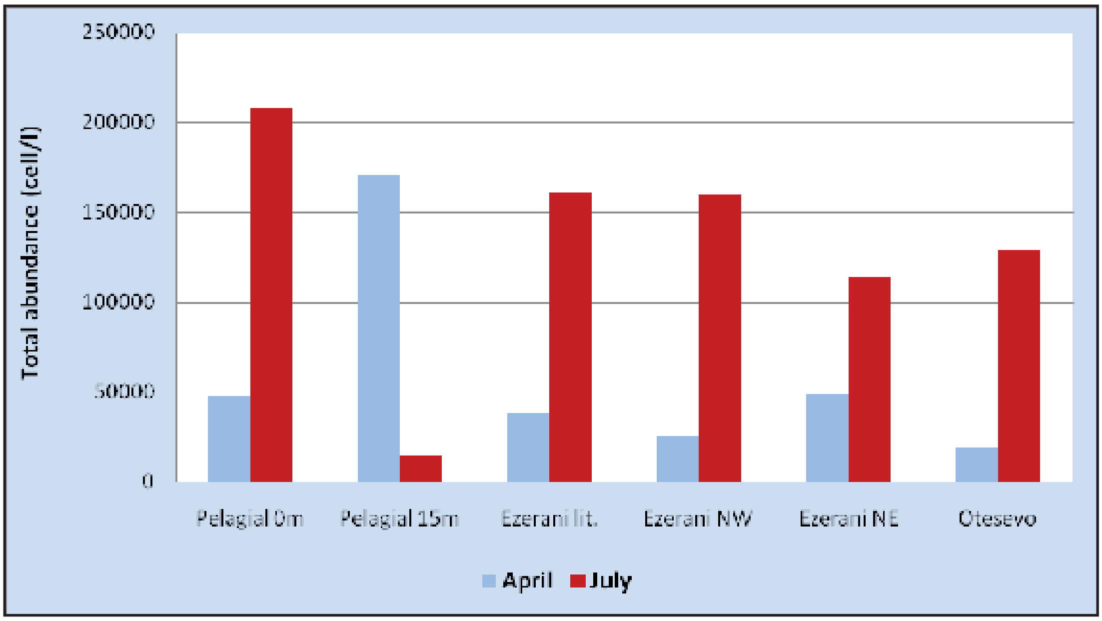
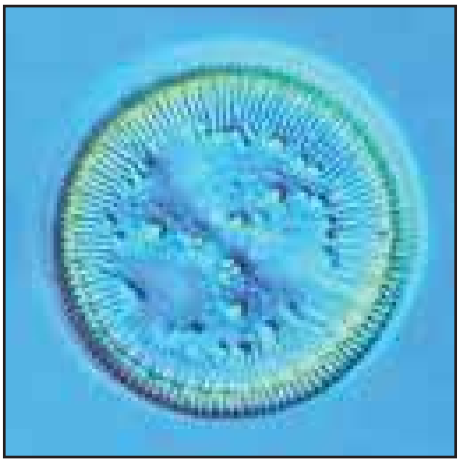


Figure 7. Phytoplankton abundance at 6 sampling points in the Macedonian part of Lake Prespa the during two sampling campaigns (cell/l)

Table. 3. Dominant species in the phytoplankton community during the two sampling campaigns in Lake Prespa

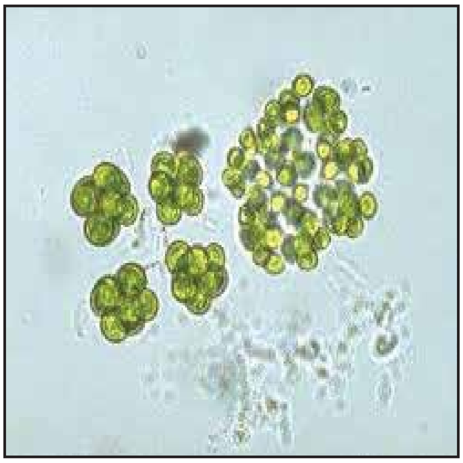
Sampling points	First (spring) sampling campaign	Second (summer) sampling campaign
Pelagial 0m	*Cyclotella ocella* Pant.	*Peridinium cunningtonii* Lemm. *Lyngbya limnetica* Lemm.
Pelagial 15m	*Cyclotella ocellata* Pant.	*Closterium acutum v. variabile* (Lemm.) Krieg.
Ezerani lit.	*Cyclotella ocellata* Pant.	*Cyclotella ocellata* Pant. *Lyngbya limnetica* Lemm.
Ezerani NW	*Cyclotella ocellata* Pant.	*Cyclotella ocellata* Pant. *Lyngbya limnetica* Lemm.
Ezerani NE	*Chlamidomonas* sp.	*Chlamiydocapsa planctonica* (West & G.S.West) Fott
Oteshevo	*Cyclotella ocellata* Pant.	*Cyclotella ocellata* Pant.



Cyclotella ocellata **Pant.**

Lyngbya limnetica **Lemm**

Peridinium cunningtonii **Lemm.**



Chlamiydocapsa planctonica
(West & G.S.West) Fott

4.4. Chlorophyll *a* concentration and Trophic State Index (TSI)

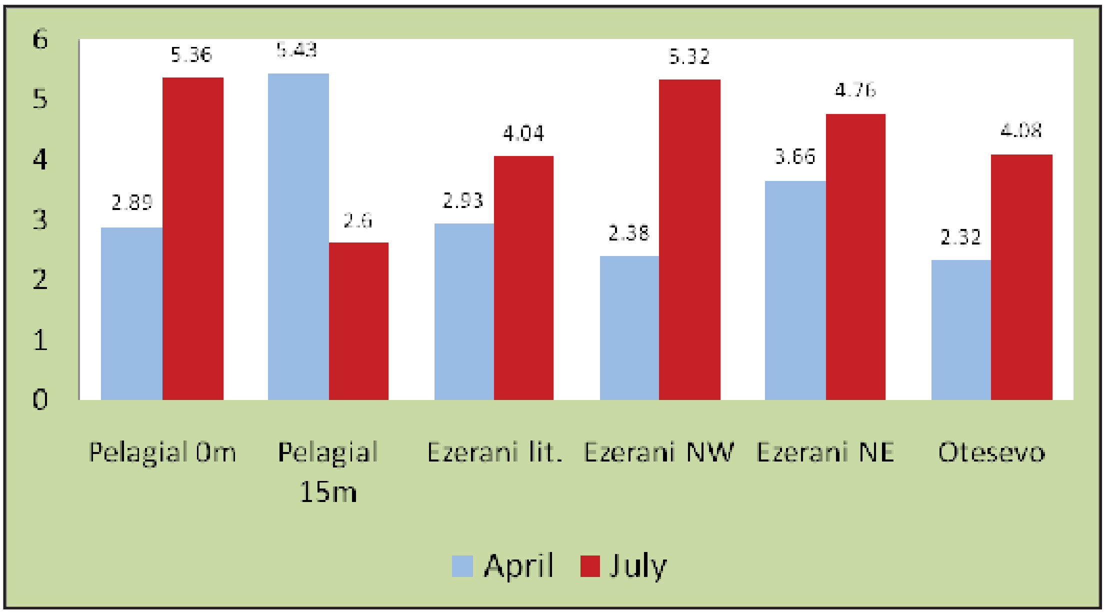


Figure 8. Chlorophyll a concentration (μg l^{-1}) in Lake Prespa

Similar to algal abundance chlorophyll *a* concentrations at surface level were generally higher in summer than in spring. The reverse was true for the pelagic zone at 15 m depth (Fig. 8).

This kind of seasonal chlorophyll *a* distribution in the pelagic zone of Lake Prespa corresponds with the typical distribution of chlorophyll *a* in mesotrophic lakes from temperate zones (Marshall and Peters, 1989).

The observed seasonal distribution of chlorophyll *a* concentrations at Lake Prespa was opposite to the one observed at Lake Ohrid, due to the different trophic states of the lakes.

Trophic State Index (TSI; Carlson, 1977) is an indicator of water trophic level. It provides the basis that links chlorophyll *a* levels and transparency with total phosphorus, which tends to fuel algal production. Calculation of this index is based on the chlorophyll *a* concentration (Chl *a*) which is an indirect measure of phytoplankton biomass:

TSI value

- Trophic level
- < 40
- Oligotrophy
- 40 - 50
- Mesotrophy
- 50 - 60
- Eutrophy I
- 60 - 80
- Eutrophy II
- > 80A
- Hypertrophy

Table. 4. Trophic state index (TSI) during the two sampling campaigns in Lake Prespa

	Pelagial 0m	Ezerani lit.	Ezerani NW	Ezerani NE	Otesevo
Spring	41	41	39	43	39
Summer	47	44	47	46	44

According to the trophic state index (TSI) all sampling points at Lake Prespa indicated mesotrophic conditions.

Table 5. Trophic state categories of lakes predicated on summer chlorophyll a concentration based on Nürnberg (1996)

Oligotrophy	Mesotrophy	Eutrophy	Hypereutrophy
< 3.5	3.5 - 9	9.1 - 25	> 25

Table 6. Trophic state based on Nürnberg (1996) during the two sampling campaigns in Lake Prespa

	Pelagial 0m	Ezerani lit.	Ezerani NW	Ezerani NE	Otesevo
Summer	5.4	4.0	5.3	4.8	4.1

According to the classification system of Nürnberg (1996) for summer periods, Lake Prespa is in mesotrophic state (Tab. 6). Using phytoplankton as a biological element according to the Water Framework Directive we conclude that the status of Lake Prespa is moderate (Tab. 7).

Table 7. Ecological Status of Lake Prespa according to WFD

Pelagial 0m	Ezerani lit.	Ezerani NW	Ezerani NE	Otesevo

5. CONCLUSIONS

Phytoplankton assemblages of Lake Prespa and their spatial and temporal distribution were found to be typical of mesotrophic lakes.

According to the Trophic State Index (TSI) all sampling sites are in a mesotrophic state.

This is also confirmed by an analysis based on the classification system of Nürnberg (1996) which considers summer Chl *a* concentrations, only.

According to the Water Framework Directive, the ecological status of Lake Prespa can be considered as **moderate**.

Bacillariophyta and Cyanophyta were the dominant divisions within phytoplankton assemblage, showing distinct seasonal variations. Bacillariophyta are predominating in spring and Cyanophyta in summer.

A distinct difference in species dominance was observed at Ezerani NE where Chlorophyta were the dominant phytoplankton group both in spring and in summer.

In summer, the total abundance of phytoplankton was significantly higher than in spring except for the pelagic zone at 15 m of depth where the reverse was observed.

Likewise, chlorophyll *a* concentrations were higher in summer than in spring, the pelagic zone at 15 m being an exception again.

Overall chlorophyll *a* concentrations were very similar among the different sampling points.

According to Prespa Lake Watershed Management Plan (GEF/UNDP) the diatom flora of Lake Prespa was very rich in taxa but the overall composition of taxa in the communities indicates an ecosystem which is naturally rich in nutrients and enables the development of diverse microflora, reflecting the basic mesotrophic state. The final support for the overall conclusion that Lake Prespa has completed the turnover to a highly eutrophic system comes from analyses of plankton communities during summer months. Only two cyanobacteria forms have produced a typical 'water bloom' from May to September which have fully replaced the usual plankton dominance of diatoms belonging to the genus Cyclotella.

In our investigations were not observed water bloom from Cyanophyta in summer. The trophic state of Lake Prespa was better than in previous last investigations.

6. RECOMMENDATIONS

According to the Water Framework Directive the status of the Macedonian water bodies of Lake Prespa can be considered as **moderate.** However, the establishment of reference conditions of water quality based on phytoplankton is difficult owing to the lack of historical and continuous monitoring schemes. Available historical data and the data generated under the current project are insufficient to derive definite conclusions.

Previous research data showed that Lake Prespa was in the oligotrophic state in the middle of the last century (Kozarov, 1959). However, in the last decades of the last century and as a result of an anthropogenic pressure, increasing eutrophication lead to the deterioration of trophic state (Patceva, 2005).

Considering that the large size of the lake and the high temporal variability of many physico-chemical parameters, phytoplankton and chlorophyll *a* parameters should be monitored on a monthly basis, especially during the summer period, with sampling points distributed over the whole lake area, because phytoplankton communities show highly specific spatial and temporal distributions in the lakes.

According to the phytoplankton parameters, the Macedonian part of Lake Prespa can be delineated as a single water body.

For future monitoring and as reference points it is strongly recommended to define sampling points at four different depths within the pelagic zone.

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Annex 1. Summary of the results of phytoplankton investigations at Lake Prespa in April and July 2013

	Sampling sites											
	Ezerani NE		Ezerani lit.		Ezerani NW		Oteshevo		Pelagic (surface)		Pelagic (15 m)	
Parameter / **Month**	**April**	**July**	**April**	**July**	**April**	**July**	**April**	**July**	**April**	**July**	**April**	**July**
Species number	11	26	16	36	6	25	8	16	6	17	6	7
Shannon diversity	0.98	2.58	1.36	2.06	0.79	2.15	0.86	1.80	0.41	2.02	0.11	1.21
Pielou's evenness	0.41	0.79	0.49	0.58	0.44	0.67	0.41	0.65	0.23	0.71	0.06	0.62
Total abundance (cells/L)	48 800	114 000	38 000	169 800	25 800	159 200	19 600	129 600	48 400	208 000	169 800	14 800
Dominant species (% of total abundance)	CO, 55											
Divisions (% of total abundance)												
– Cyanophyta	1	21	1	35	0	34	0	23	3	31	1	18
– Bacillariophyta	8	20	76	40	81	36	84	41	93	12	98	15
– Chlorophyta	84	44	17	21	18	19	14	15	4	21	1	67
– Chrysophyta	0	0	0	1	0	9	2	0	0	1	0	0
– Pyrrophyta	7	12	6	2	1	1	0	21	0	35	0	0
– Euglenophyta	0	3	0	1	0	1	0	0	0	0	0	0
Chlorophyll a concentration (µg/L) [1]	3.7	4.8	2.9	4.0	2.4	5.3	2.3	4.1	2.9	5.4	5.4	2.6
TDI (based on chlorophyll a concentration)	43	46	41	44	39	47	39	44	41	47	-	-

[1] Color according to summer (July) Chlorophyll a concentrations (µg/L) following Nürnberg (1996)

Annex 2. List of identified phytoplankton species at Lake Prespa in April and July 2013

		Pelagial 0m	Pelagial 15m	Ezerani NW	Ezerani lit.	Ezerani NE	Oteshevo
	CYANOPHYTA						
1.	Anabaena flos-aquae Bréb.	+		+			
2.	Anabaena solitaria fo. planktonica (Brunn.) Komárek	+		+	+	+	+
3.	Aphanizomenon flos-aquae (L.) Ralfs	+					
4.	Chroococcus limneticus Lemm.	+		+	+	+	+
5.	Chroococcus minimus (Keissl.) Lemm.				+	+	+
6.	Lyngbya limnetica Lemm.	+	+	+	+	+	+
7.	Merismopedia glauca (Ehr.) Näg.				+		
8.	Gomphosphaeria lacustris Chod.	+			+		
	BACILLARIOPHYTA						
9.	Cyclotella sp.	+	+	+	+	+	+
10.	Cyclotella ocellata Pant.	+	+	+	+	+	+
11.	Amphora ovalis Kütz.					+	
12.	Aulacoseira sp.		+				
13.	Navicula **sp.**			+	+	+	+
14.	Fragilaria crotonensis Kitton	+					
15.	Fragilaria ulna (Nitz.) Lange-Bertalot				+		
16.	Fragilaria sp.		+	+	+	+	+
17.	Cocconeis pediculus Ehr.			+	+		
18.	Gomphonema sp.				+		
19.	Stauroneis sp.				+		
20.	Cymbella sp.				+		
	CHLOROPHYTA						
21.	Ankistrodesmus falcatus (Corda) Ralfs					+	
22.	Ankistrodesmus lacustris Ostenf.		+	+	+	+	
23.	Chlamidomonas sp.	+		+	+	+	+
24.	Closterium acutum v. variabile Krieger	+	+	+	+	+	+
25.	Cosmarium phaseolus Bréb.				+		
26.	Staurastrum paradoxum Meyen				+		
27.	Staurastrum paxilliferum G. S. West				+		+
28.	Oocystis lacustris Chod.	+	+	+	+	+	+
29.	Oocystis rhomboides Fott	+		+	+	+	+
30.	Chlamidocapsa planctonica	+	+	+	+	+	+
31.	Scenedesmus quadricauda Bréb.				+	+	
32.	Scenedesmus obliquus (Turpin) Kütz.			+	+		+
33.	Scenedesmus acuminatus (Lag.) Chod.				+	+	

		Pelagial 0m	Pelagial 15m	Ezerani NW	Ezerani lit.	Ezerani NE	Oteshevo
34.	Scenedesmus arcuatus Lemm.				+		
35.	Pediastrum boryanum (Turp.) Mengh.					+	
36.	Pediastrum duplex Meyen			+	+		
37.	Pediastrum simplex Meyen				+	+	
38.	Pandorina morum (Muell.) Bory			+	+	+	+
39.	Eudorina elegans Ehr.			+	+	+	
40.	Nephrocytium aghardhianum Nägel						
41.	Dictyosphaerium pulchelum Wood.			+	+	+	
42.	Sphaerocystis shroeterii Chod.					+	+
43.	Mougeotia sp.				+		+
44.	Spirogyra sp.						+
	CHRYSOPHYTA						
45.	Dinobryon divergens Imhof				+		
46.	Dinobryon sociale v. stipitatum Lemm.			+	+	+	
47.	Dinobryon bavaricum Imhof				+		
	PYRROPHYTA						
48.	Gymnodinium mirabile var. rufescens Penard.			+			
49.	Gymnodinium sp.	+		+	+	+	
50.	Peridinium cunningtonii Lemm.	+		+	+	+	+
51.	Peridinium cinctum (O. F. Müll.) Her				+	+	
52.	Peridinium sp.					+	+
	EUGLENOPHYTA						
53.	Euglena viridis Ehr.			+			
54.	Euglena sp.				+	+	
55.	Phacus sp.			+	+	+	
56.	Trachelomonas sp.				+	+	

PHYTOPLANKTON OF MACRO PRESPA LAKE
(Albanian part)
APRIL 2014

Marash Rakaj
Water Research Institute of Shkodra Region,
University of Shkodra "Luigj Gurakuqi", Shkoder

Shkoder, April 2014

Contents

1. Introduction

Phytoplankton as primary producers have a primary role in aquatic food webs and an important influence on other organisms. They are one of several biological quality elements (BQE) used to assess the ecological status of water bodies according to the European Union Water Framework Directive (WFD). The element is very dynamic and strongly dependent on, nutrient concentrations, light intensity, temperature and other environmental factors.

Due to their short life cycle, phytoplankton responds quickly to environmental changes and is thus a valuable indicator of water quality.

The present study investigates the phytoplankton of Macro-Prespa according to an agreed CSBL sampling protocol, focusing on the following parameters:

- **Phytoplankton composition**
- **Species abundance**
- **Total abundance**
- **Trophic State Index**

Phytoplankton are increasingly used to assess the ecological quality and status of the water environment and to measure and monitor the effectiveness of management or restoration programmes or regulatory actions.

Objectives:

- **Sample phytoplankton at two selected sampling points of Macro-Prespa**
- **Conduct species inventory (qualitative composition of phytoplankton assemblages).**
- **Assess phytoplankton abundance (enumeration) of recorded species (quantitative composition of phytoplankton)**
- **Calculate the Trophic State Index (TSI) according to Carlson (1977) based on transparency or Secchi disc values and Chlorophyll a (the latter being determined in parallel by the University of Tirana).**

2. Parameters and sampling points

Phytoplankton samples were taken on April 4 at two sampling points (**Gollomboc and Pustec**) at the littoral part of Macro Prespa (Figure 1) and analyzed according to a CSBL phytoplankton protocol prepared by Jelena Rakočević in 2013.

The most common physico-chemical parameters such as **temperature (°C), transparency, pH** and **dissolved oxygen** were measured (Table 1). Water samples of up to 0.25literwere taken from a boat at 0.5m depth using a Van Dorm bottle (Figure2).

Figure 1. Phytoplankton sampling points at the Albanian part of Macro Prespa Lake

Figure 2. Sampling in Pustec

Table 1. Coordinates and physico-chemical parameters of sampling points

Nr.	Sampling Points	Coordinates N	E	T (°C)	Transparency (m)	pH	Dissolved O_2 (mg/l)
1.	Gollomboc	40^0 51' 43.8"	20^0 56' 25.4"	10.7	3.7	7.8	8.2
2.	Pustec	40^0 47' 27.2"	20^0 54' 42.2"	10.8	3.4	8.1	7.8

3. Storage of samples and transport

Samples for counting were preserved immediately at the sampling site by adding *Lugol's solution* (preserve 1 L-sample with 3 ml of *Lugol's solution*) while samples for species determination were preserved with *formaldehyde* (0.4%).

Each sample was clearly labeled indicating sampling site as well as date, time and depth of sampling.

Sample bottles were transported to the laboratory in a cooler box and were then stored in a dark place (shelf).

4. Analytical methods

Qualitative and quantitative analysis of phytoplankton was done according to standard methods (Utermöhl, 1958; ISO 10260, 1992; Carlson, 1996) and CSBL – Protocol for phytoplankton (Table 2).

Table. 2. Method used in monitoring program implementation for Macro Prespa Lake

Variable: Phytoplankton Communities

No.	Method	Equipment	Sampling method/container	Preservation
1.	Utermöhl (1958)	Microscope Optic Leitz-Diaplan	Van Dorm bottle 2l	Formaldehyde 4%
		Inversion Microscope 200-400x (Axiovert 25),	Plankton net, Secchi disc	Lugol 1%
2.	Carlson(1977, 1996)	Utermöhl chambers	Glass bottles 250ml	

Identification of species of phytoplankton was done using a microscope Leitz-diaplan with immersion lenses 63/1.4 Plan APO according to Bourrely (1970, 1981), Komarek & Anagnostidis (1999), Krammer & Lange-Bertalot (1986, 1988, 1990), Pestalozzi, Kramer & Fott (1983) and Popovsky & Pfiester (1990).

Quantitative analysis or enumeration of individuals was done at chamber 20ml using an inverted microscope at up to 400-xmagnification (Axiovert 25)

Diversity index was calculated according to Shannon-Wiener (1949) and evenness according to Pielou (1975).

The Trophic State Index (TSI) was calculated according to Carlson (1977), based on transparency (Secchi Disc).

5. Results and evaluation

A total of30 species were identified, representing 5 phytoplankton groups: Bacillariophyceae – 18 species or 60%, Chlorophyceae – 4 species or 13.33%, Cyanophyceae – 3 species or 10%, Dinophyceae (Pyrrophyta) – 3 species or 10% and Chrysophyceae – 2 species or 6.7% (Table 3 and Figure 3).

Table 3. Phytoplankton communities or species richness on Macro Prespa (April 2014)

Nr.	Groups/Divisions	Gollomboc	Pustec	Total species	Total genera
1	Chrysophyceae	2	2	2 (6.7%)	1
2	Bacillariophyceae (Diatoms)	14	16	18 (60.0%)	13
	Centrales	3	3	3	2
	Pennales	11	13	15	11
3	Dinophyceae (Peridine)	3	3	3 (10.0%)	2
4	Chlorophyceae	3	4	4 (13.3%)	4
5	Cyanobacteria	2	3	3(10.0%)	3
	Total	**24**	**28**	**30**	**23**

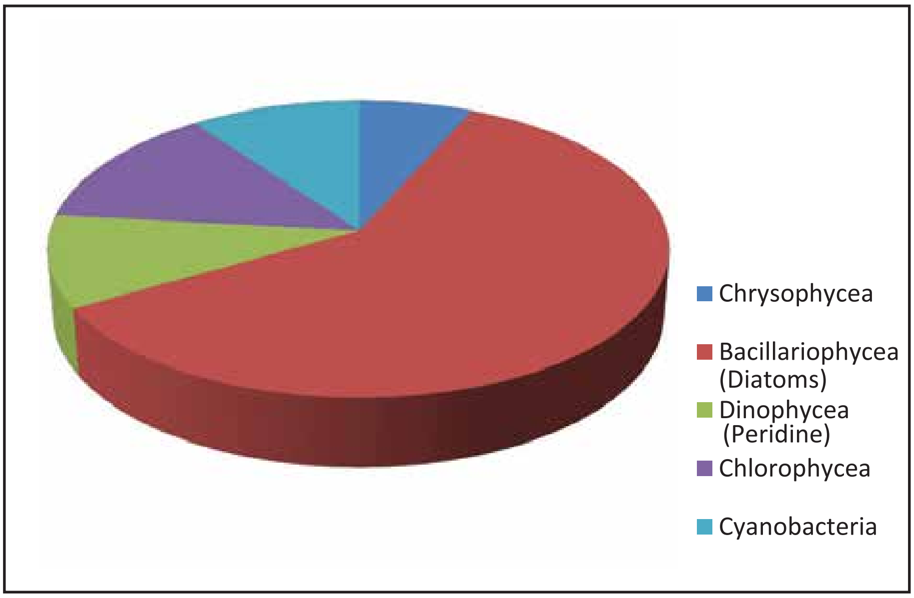

Figure 3. Phytoplankton communities of Macro Prespa Lake in April, 2014

Phytoplankton assemblages were dominated by diatoms (Bacillariophyta) which we represented with 18 species or 60% of the total number of species. Other groups were represented by smaller numbers of species: 4 Chlorophyta, 3 Pyrrophyta, 3 Cyanophyta and 2 species Chrysophyceae. Phytoplankton abundance at both sampling points was dominated by diatoms, mainly *Cyclotella ocellata, C. fotti*, *Diploneis mauleri, Nitschia subacicularis* and *Campylodiscus noricus*. The second most dominant group was Dinophyceae, represented mainly by *Gymnodinium mirabile*, and Chrysophyceae, represented by only one genus, *Dinobryon*. *Chlorophyta* (mainly Chlorococcales) and Cyanobacteria were found less often.

The distribution of species at the two sampling points was homogenous. Twenty-eight species were found at Pustec and 24 species at Gollomboc. The density or number of individuals per liter water was slightly higher in Pustec ($2.84x10^4$) than in Gollomboc ($2.19x10^4$).

Diversity and **Evenness** were higher for Gollomboc (2.34 and 0.68) than for Pustec (2.13 and 0.62; Table 4).

The higher indices at Gollomboc were due to lower and more uniform abundances of phytoplankton species at this sampling site.

Table 4. Diversity and evenness of phytoplankton assemblages of Macro Prespa

Nr.	Parameters/Stations	Gollomboc	Pustec
1.	**Species number (S)**	24	28
2.	**Diversity index (H')** $H' = -\sum_{i=1}^{R} p_i \ln p_i$	2.34	2.13
3.	**Evenness index** ($J' = H'/H'_{max}$ or H'/lnS)	0.68	0.62

As shown in Table 5, the highest **total abundance** of phytoplankton was recorded at Pustec ($2.84x10^4$) and the lowest at Gollomboc ($2.19x10^4$). The higher abundance at station of Pustec compared to Gollomboc supposedly reflects higher nutrient concentrations.

Table 5. Phytoplankton abundance (cells/l) of different groups at Macro Prespa

No.	Groups/sampling points	Gollomboc	Pustec
1	**Chrysophyceae**	3.18×10^3	3.27×10^3
2	**Bacillariophyceae (diatoms)**	12.81×10^3	14.95×10^3
3	**Dinophyceae (Peridine)**	6.70×10^3	7.80×10^3
4	**Chlorophyceae**	4.80×10^2	5.60×10^2
5	**Cyanobacteria**	1.10×10^2	2.12×10^2
	Total density	2.3×10^4	2.7×10^4

Dominant species were diatoms, including the *Cyclotella ocellata* complex with up to 11.60×10^3cells/l, associated with *Diploneis mauleri* (Fig. 5a), *Campylodiscus noricus* and *Nitschia subacicularis*. The second and third most abundant groups were Dinophyceae (Pyrrophyta) represented by *Gymnodinium mirabile* with $6.9 \times 10^3$and Chrysophyceae represented by *Dinobryon* spp., while other groups were presented by only few species and much lower abundance. Two filamentous species (*Anabaena, Planktolyngbya*; Fig. 5b,c) were present at the surface water of Pustec.

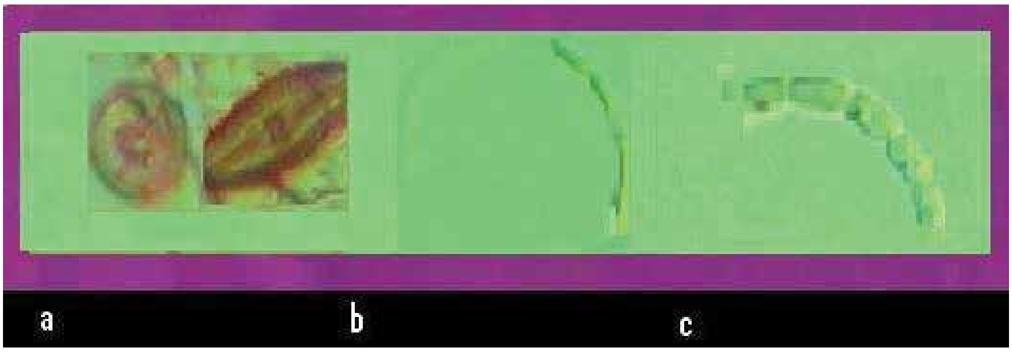

Figure 5. a – *Cyclotella ocellata* **&** *Diploneis mauleri*;
b– *Planktolyngbya contorta*; **c**– *Anabaena contorta*

The **Trophic State Index (TSI)** was calculated based on values of water transparency measured with a Secchi disc according to **TSI(SD)** = 60 - 14.41 ln(SD)(Table 6).

Table. 6. TSI values based on SD transparency measurement

No.	Stations	**Transparency** (m)	**Trophic State Index** (SD)
1.	**Gollomboc**	3.7	41
3.	**Pustec**	3.4	42

The relatively low abundance of phytoplankton (2.3×10^4 and 2.7×10^4cells/l)at this time of the year suggests oligotrophic conditions while the TSI(SD) values > 40 indicate mesotrophic conditions at both sampling sites.

There appeared to be no clear relationship between values TSI and phytoplankton abundance at either site (Table 6). The values of TSI are slightly higher probably because of unstable water condition at the littoral part of the lake.

The slightly higher phytoplankton abundance and TSI at Pustec compared to Gollomboc appears to be due to higher nutrient levels as a result of more intensive agricultural activities at Pustec and Zaroshke.

The overall low abundance (biomass) of phytoplankton presumably results from uncompleted stabilization of water conditions and low water temperature at the time of sampling.

6. Summary and Recommendations

Field investigations at two sampling sites in April 2014 revealed 30 species of five different phytoplankton groups. Bacillariophyceae(diatoms) were the most species-rich group, comprising 18 species or 60% of the total species number.

Within the Bacillariophyta, species such as *Cyclotella ocellata, C. fotti, Diploneis mauleri, Campylodiscus noricus* and *Nitschia subacicularis* were the most abundant. The second dominant groups were Dinophyta represented mainly by *Gymnodinium mirabile* and Chrysophyceae, with two species of the genus *Dinobryon*.

The values of phytoplankton abundance suggest oligotrophic conditions at both sampling points, even though the abundance at Pustec as slightly elevated ($2.8x10^4$ cells/l). The TDI in contrast, suggest slightly mesotrophic conditions. However, the TSI based on SD depth measurements is only a proxy for phytoplankton biomass and has more limited significance regarding the assessment of trophic state compared to the TSI based on Chlorophyll a. For the latter, see the results of the chemotaxonomic analysis done in parallel to the present study

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8. **Annexes** *(attached in Excel format)*

1. Taxonomic list of species for both sampling points (stations)

TAXONOMIC LIST AND SPECIES FREQUENCY ON BOTH STATION

Groups	Scientific names/Stations	Gollomboc	Pustec
Chrysophycea	*Dinobryon bavaricum*	1	2
	Dinobryon divergens	1	1
Bacillariophycea (Diatoms)			
Centrales	*Cyclotella ocellata*	3	3
	Cyclotella fotti	2	2
	Campylodiscus noricus	2	2
Pennales	*Amphora ovalis*	a	1
	Cymbella ehrenbergii	1	2
	Cymbella lanceolata	1	1
	Cymatopleura elliptica	1	2
	Diploneis mauleri	2	3
	Epithemia turgida	1	a
	Frgilaria crotonensis	a	2
	Gomphonema truncatum	1	1
	Gyrosigma attenuatum	1	1
	Navicula hastata	2	2
	Navicula rotunda	a	1
	Navicula (Cavinula) scutelloid	1	2
	Nitzschia sigmoidea	a	1
	Nitzschia subacicularis	2	3
	Synedra acus	1	2
Dinophycea (Peridine)	*Gymnodinium mirabile*	1	2
	Peridinium cinctum	1	2
	Peridinium cuningtoni	2	2
Chlorophycea	*Ankistrodesmus lacustris*	1	1
	Scenedesmus quadricauda	1	1
	Oocystic lacustris	2	1
	Closterium setaceum	a	1
Cyanobacteria	*Chroococcus limneticus*	1	2
	Anabaena contorta	a	1
	Planctolyngbia contorta	1	2
a = absent			

2. Phytoplankton communities or species richness

Phytoplankton communities or species richness on April 2014						
Nr.	Groups/communities	Gollombac	Pustec		Total species	Total genera
1	Chrysophycea	2	2		2 (6.66%)	1
2	Bacillariophycea (Diatoms)	14	16		18 (60%)	13
	Centrales	3	3		3	2
	Pennales	11	13		15	11
3	Dinophycea (Peridine)	3	3		3 (10%)	2
4	Chlorophycea	3	4		4 (13.3%)	4
5	Cyanobacteria	2	3		3(10%)	3
	Total species	**24**	**28**		**30**	**23**

3. Abundance of phytoplankton on both stations

ABUNDANCE OF PHYTOPLANKTON (cell/l) ON BOTH STATIONS			
Nr.	Groups/sampling points	Gollomboc	Pustec
1	Chrysophycea		
	Dinobryon sp.div	$3.18x10^3$	$3.27 x10^3$
2	Bacillariophycea (Diatoms)		
	Cyclotella	$9.01 x10^3$	$11.30 x10^3$
	Diploneis, Campilodiscus and .	$3.80 x10^3$	$3.65 x10^3$
3	Dinophycea (Peridine)		
	Gymnodinium mirabile	$5.9x10^3$	$6.9x10^3$
	Peridinium sp.div	100-300	100-300
4	Chlorophycea		
	(mainly Chlorococcales)	$4.8x10^2$	$5.60x10^2$
5	Cyanobacteria		
	Planctolybya limnetica	10-100	100-300
	Total density	$2.3x10^4$	$2.7x10^4$

4. Summary of phytoplankton abundance

SUMMERY OF PHYTOPLANKTON ABUNDANCE (cell/l)			
	Groups/sampling points	Gollomboc	Pustec
1	Chrysophyceae	$3.18x10^3$	$3.27x10^3$
2	Bacillariophyceae (Diatoms)	$12.81x10^3$	$14.95x10^3$
3	Dinophyceae (Peridine)	$6.70x10^3$	$7.80x10^3$
4	Chlorophyceae	$4.80x10^2$	$5.60x10^2$
5	Cyanobacteriae	$1.10x10^2$	$2.12x10^2$
	Total density	$2.3x10^4$	$2.7x10^4$

5. Quantitative data for both stations

Quantitative Data by different sampling points				
Nr.	Sampling points	Eveness Index (J')	Abundace	Trophic State Index (TSISD)
1	Gollomboc	0,68	$2.19x10^4$	41,15
2	Pustec	0,62	$2.84x10^4$	42,37

6. Calculation of different indexes.

CALCULATION Of DIFFERENT INDEXES			
I.	DIVERSITY INDICES (H) OF BOTH STATIONS		
Sampling point/1	$H' = -\sum_{i=1}^{R} p_i \ln p_i$	H = 2.34	
2		H = 2.13	
	pi=ni/N		
II.	EVENESS INDEX OF THREE DIFFERENT STATIONS		
	$J' = H / H'_{max}$	H'max = lnS	
Sampling point 1	S = 30	J = 2.34/ln30	J = 0.68
Sampling point 2		J = 2.13/ln30	J = 0.62

Annex 1

RESULTS OF PHYTOPLANKTON ANALYSES

Date 04.04.2014

Table 1. Coordinates and physico-chemical parameters of stations of Macro Prespa on April 4th

Nr.	Sampling Points	Coordinates N	E	T (°C)	Transparency (m)	pH	Dissolved Oxygen (mg/l)
1	Gollomboc	40^0 51' 43.8"	20^0 56' 25.4"	10,7	3,7	7,8	8,2
2	Pustec	40^0 47' 27.2"	20^0 54' 42.2"	10,8	3,4	8,1	7,8

Annex 2

TAXONOMIC LIST AND SPECIES FREQUENCY ON BOTH STATION

Groups	Scientific names/Stations	Gollomboc	Pustec
Chrysophycea	*Dinobryon bavaricum*	1	2
	Dinobryon divergens	1	1
Bacillariophycea (Diatoms)			
Centrales	*Cyclotella ocellata*	3	3
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	Campylodiscus noricus	2	2
Pennales	*Amphora ovalis*	a	1
	Cymbella ehrenbergii	1	2
	Cymbella lanceolata	1	1
	Cymatopleura elliptica	1	2
	Diploneis mauleri	2	3
	Epithemia turgida	1	a
	Frgilaria crotonensis	a	2
	Gomphonema truncatum	1	1
	Gyrosigma attenuatum	1	1
	Navicula hastata	2	2
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	Scenedesmus quadricauda	1	1
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	Closterium setaceum	a	1
Cyanobacteria	*Chroococcus limneticus*	1	2
	Anabaena contorta	a	1
	Planctolyngbia contorta	1	2

a = absent

Composition and Abundance of Benthic Invertebrates (Macrozoobenthos) at Selected Sites of the Albanian Territories of Lake Prespa[1]

Prof Dr Sajmir Beqiraj

Department of Biology

Faculty of Natural Sciences

University of Tirana

Tirana, 18th November 2014

1 Summary report compiled by CSBL based on a presentation given by the author.

Contents

Biological quality elements – Benthic invertebrate fauna

1. Macroinvertebrate communities

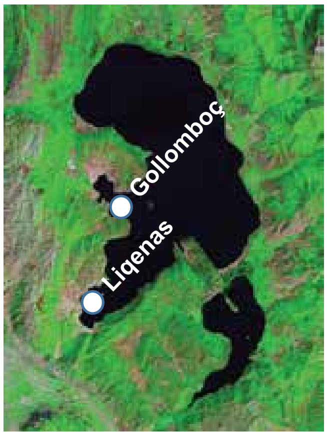
Macroinvertebrate communities are:

- used as very good and accurate biological indicators for the ecological status of lakes
- show different tolerances to nutrients, pollutants, siltation, low oxygen levels and modifications of substrate or hydromorphology
- have rarely been studied at the Albanian part of Prespa Lake
- recommended to be monitored at least every 3 years for water bodies at risk of failing to achieve good status (under the operational monitoring scheme)






2. Sampling period and methodology



- Sampling period: October 2013 and May 2014
- Sampling sites: Gollomboç and Liqenas
- Sampling methodology (ISO: EN 27828: 1994):
  - Kick and swipe for the shallow part (0.5 m depth)
  - Multi-habitat transect method (2, 4, 6 and 10 m depth)



3. Ecological status of Prespa Lake, based on benthic macroinvertebrates as bio-indicators

Table 1 Macroinvertebrate Biotic Index (MBI), Biological Monitoring Work Party (BMWP) index and Average Score Per Taxon (ASPT) index at different water depths at Gollomboç.

Depth (m)	MBI	Quality	BMWP	Quality	ASPT	Quality
0.5	6.0	Poor	36.0	Poor	3.6	Bad
2	6.1	Poor	30.4	Poor	3.8	Poor
4	6.0	Poor	19.3	Poor	3.2	Bad
6	6.0	Poor	25.6	Poor	3.2	Bad
10	6.0	Poor	16.6	Poor	4.1	Poor

Table 2 MBI, BMWP and ASPT at different water depths at Liqenas.

Depth (m)	MBI	Quality	BMWP	Quality	ASPT	Quality
0.5	6.0	Poor	29.3	Poor	4.1	Poor
2	6.1	Poor	12.6	Bad	3.1	Bad
4	6.1	Poor	16.1	Poor	3.2	Bad
6	6.0	Poor	9.7	Bad	3.2	Bad
10	6.1	Poor	13.9	Bad	3.4	Bad

Table 3 Community structure assessment, based on different diversity indices at different water depths at Gollomboç.

Depth (m)	Shannon	Status	Pielou	Status	Margalef	Status	Simpson (D)	Status
0.5	2.245	Moderate	0.697	Moderate	3.453	I	0.135	High
2	1.848	Moderate	0.667	Moderate	2.862	II	0.217	Good
4	1.187	Poor	0.463	Poor	2.234	IV	0.406	Poor
6	1.591	Moderate	0.588	Poor	2.647	III	0.276	Good
10	1.159	Poor	0.720	Moderate	1.005	V	0.354	Moderate

Table 4 Community structure assessment, based on different diversity indices at different water depths at Liqenas.

Depth (m)	Shannon	Status	Pielou	Status	Margalef	Status	Simpson (D)	Status
0.5	1.530	Moderate	0.596	Poor	2.136	II	0.292	Good
2	1.457	Poor	0.525	Poor	2.561	I	0.353	Moderate
4	1.375	Poor	0.661	Moderate	1.574	III	0.321	Moderate
6	1.274	Poor	0.655	Moderate	1.163	V	0.329	Moderate
10	1.338	Poor	0.643	Moderate	1.474	IV	0.297	Good

4. Conclusions

- Macrozoobenthic communities in the Albanian part of Lake Prespa are characterized by relatively low species richness, low abundance and degradation of the population structure.
- A slightly better ecological state has been recorded in spring season (not shown).
- The environmental quality of the lake is predominated by the 'poor' to 'bad' status, which is more pronounced at Liqenas and in the fall season.
- Indicator benthic macroinvertebrates reflect a tendency for lake eutrophication, enrichment in nutrients and increased organic pollution.
- However, the lake is (still) a shelter for many benthic macroinvertebrate species of international concern and conservation interest.

Fish Monitoring and Fisheries

Lake Prespa

Final Report

July 2014

Lead authors:

Dusica Ilik-Boeva – HIO

Zoran Spirkovski – HIO

Co-authors:

Spase Sumka – PPNEA/AUT

Trajce Talevski - HIO

Technicians:

Kosta Trajce, Pellumb Aleksi, Olsi Duma – FMO Prespa

Gjorgi Puftoski, Zaim Ramadan - HIO

Student volunteer:

Blagoja Trajcevski - HIO

Contents

1. Summary

The Project Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar regarding fish and fisheries component gave in some cases improved knowledge about Lake Prespa fish stock. In this case, the sampling campaigns in the autumn period gives main contribution for the ecological moments of the alien species correlated with the habitats, and regarding the commercially valued fish species very little knowledge improvement was achieved.

The Project Conservation and Sustainable Use of Biodiversity at Lakes Prespa, Ohrid and Shkodra/Skadar regarding Lake Prespa fish fauna gives a good exercise of possibilities to evaluate the implementation of EN 14757 MMG but in such large water body using sampling procedure based on stratified random sampling of fish assemblages out of their aggregation periods as spawning and winter schooling can give only partial knowledge about ecological status of fish population and the ecological status of the water body.

From 128 randomly set MMGN in the 2013 sampling campaign 15 fish species were detected on Albanian side and 12 fish species on Macedonian side. The most abundant species on both sampling sides were bitterling (*Rhodeus amarus*) and stone moroko (*Pseudorasbora parva*).

Generally, introduced species (bitterling and stone morocco) and native *Alburnoides prespensis* (spirling) with smaller body size have high abundance in the strata 0-3 meters, while in the deeper strata species with bigger body size both native *Alburnus belvica* (bleak) and *Rutilus prespensis* (roach) and introduced *Lepomis gibbosus* (pumpkinseed) are most abundant.

In all littoral sub-basins bitterling was the predominant species in number of individuals (with 48% at SB1, 54% at SB2, 35% at SB3, 66% in SB4 and 45% in SB5) except for SB3 where the stone moroko was predominant with 43%. In the pelagic SB6 bleak is the most abundant species with 50% followed by the roach with 38%.

Species biodiversity is affected by pressures:

- The main extraction of the fish as biomass is by the fish eating birds (cormorants and pelicans) and fisheries.
- Main eutrophication source in the Prespa Lakes watershed is agriculture with high nutrient load and other harmful substances.
- High number of alien fish species is another pressure to the native fishes due to the spawning grounds inter-competition and overlap of the ecological niche.
- Fluctuation of the Prespa Lake water level is a result of changed hydrology conditions starting from 1987 and exploitation of its water for agricultural needs in the region.

Application of the performed sampling scheme and methodology provide us some basic needs regarding WFD, which can be improved by widening it in fishing gears, other fish surveillance devices and time series.

2. Introduction

The aim of the CSBL project is to improve the implementation of legislation, regulations and management plans for the conservation and sustainable use of biodiversity at lakes Prespa, Ohrid and Shkodra/Skadar. Component 2 of the CSBL plan of operation is targeting on fish monitoring and fisheries. Main tasks within this component are the implementation of joint monitoring for Lakes Prespa, Ohrid and Shkodra/Skadar, human, institutional and organizational capacity building.

This report concerns only the results, preliminary conclusions and recommendations on the fish stock stage at Lake Prespa – based on the fish monitoring by multi mesh size fishing standards and existing fishery statistics.

The agreed monitoring scheme is based on multi-mesh gillnetting according to the European Standard

EN 147 57, as a central and common sampling element in all lakes. This kind of sampling is practiced for the first time on Lake Prespa.

The requirements of the WFD in relation to fish communities are in assessing population status from species composition, abundance and age classes which in this case can partially contribute to the assessment of the current ecological state of Lake Prespa.

3. Fish fauna and fisheries at Lake Prespa

The Prespa Lakes Basin is a high altitude system (850-2600 m) with a catchment area of over 2.500 km^2; it covers parts of the territories of Albania, Macedonia and Greece. Macro Prespa has a surface area of 285 km2 with a maximum water depth of 54 m. Micro Prespa has a much smaller surface area of 47 km2 and is shallower with maximum water depth of 9 m. This lake is almost entirely situated in Greek territory except for a smaller area within Albania. Macro Prespa has four tributaries: Agios Germanos (Greek part), Brajcinska, Kranska and Golema Reka (Macedonian part). In the Albanian part there are no perennial streams.

The basin is home to nearly 30.000 people with the majority residing in Macedonia. The Region has extensive industry and the main source of income is agriculture which is estimated to employ about 75% of the work-force.

As a result of the agricultural impact, since the late 20th century the ecosystem of the Prespa Lakes has been subject to the dramatic over-abundant plant growth due to excessive nutrients (eutrophication). In the same time, reduced water levels due to over-exploitation for irrigation; and climate change made caused significant changes in the aquatic and environmental parameters of the lakes. The water transparency of the lakes, for example, is now 30-70 per cent lower than it was only fifteen years ago.

The average winter water temperatures have decreased by approximately 4^{o}C over the last twenty years as a result of reductions in the water level. This, in turn, has led to the freezing over of the lakes' littoral zones in winter. The dissolved oxygen concentrations now found in the Prespa lakes are typical of eutrophic lakes. The presence of anoxia (reduced oxygen) in the water column below 15 m is a regular phenomenon during the stagnant summer period.

The ecosystem and the biodiversity of the Region are worth special mention. The geography, soil types and climate coupled with the relatively low human population and impact in the basin has resulted in high species diversity with significant proportion of endemic species.

One National Park in Albania (Prespa) and two in Macedonia (Galicica and Pelister) are areas under protection of biodiversity.

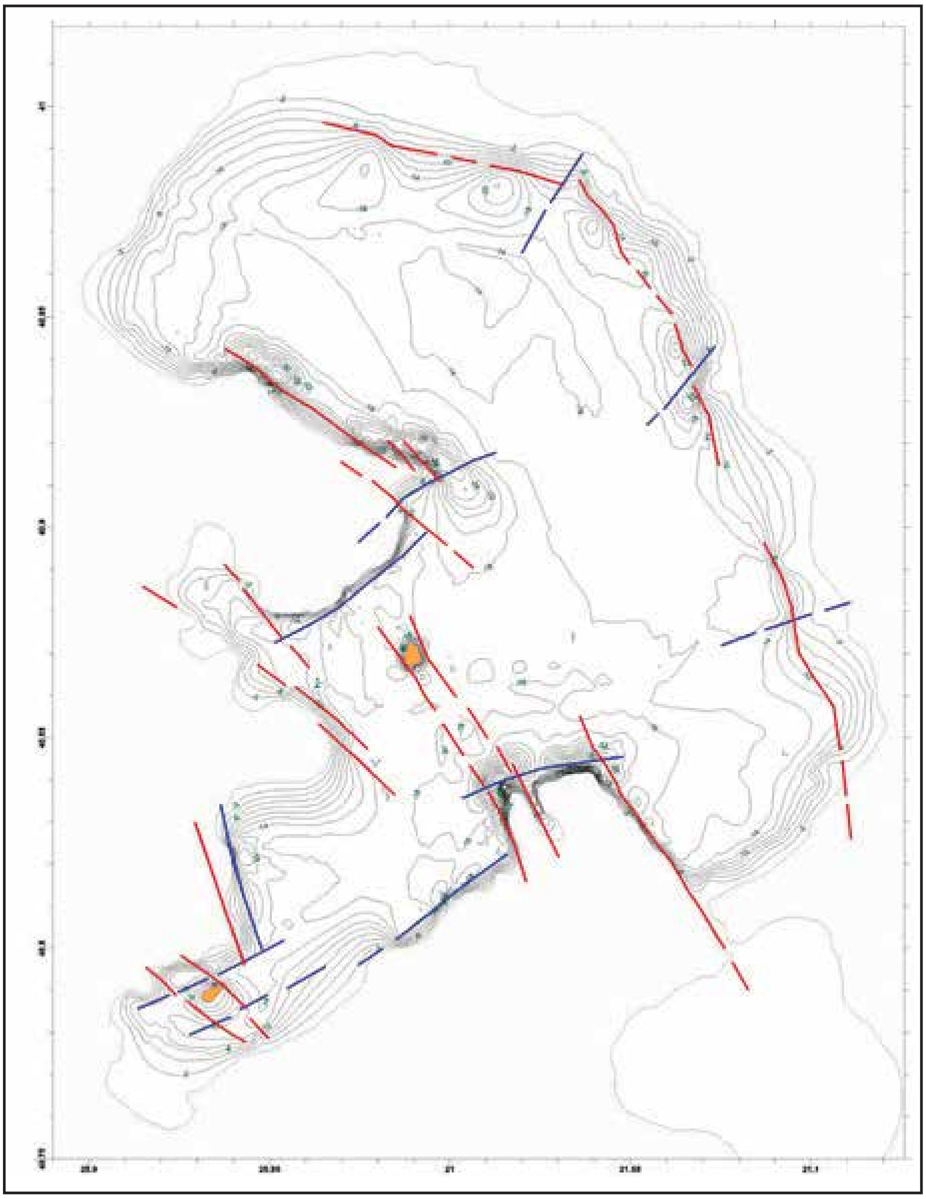


Figure 1. Tectonic and bathymetric map of Macro Prespa Lake (Source: Traborema Final report, 2007)

Lake Prespa is a subtropical dimictic lake. Changes in the trophic state of the lake are recorded in the last century. From oligotrophic until mid-70's to mesotrophic in the following two decades and eutrophic from the turn of the century. All changes in the lakes surface and volume in the last three decades are resulting increased internal pressure of the water to the sediments, and the decrease of the oxygen in the hypolimnion leads to a bigger solubility of the inorganic phosphate compounds in the water column of the Lake.

Both physico-chemical parameters and phytoplankton are showing that Prespa Lake is in the process of eutrophication, suffering dramatic changes. (Prespa Lake Fishery Master Plan, 2011)

3.1. **Fish fauna and stocks**

The Lake Prespa region has been recognized as a European and Global Hotspot of Biodiversity, not only because of the sheer number of species and habitats present, but also due to their quality, such as rarity and conservation significance. The most striking feature of the Prespa Lake Watersheds' Biodiversity is its enormous richness and heterogeneity. The total number of animal species, recorded in Macedonia's part of Lake Prespa watershed is over 2.500, of which 375 are vertebrates. Twenty-five taxa have been identified in the Prespa Lakes. None of them is migratory species.

Table 1. Lake Prespa fish species (native and alien)

Species Latin name	Species common name	native species	alien species (year of introduction)	alien species recorded in the past
Alburnoides prespensis	Prespa spirlin	+		
Alburnus belvica	Prespa bleak	+		
Anguila anguila	European eel	+		
Barbatula sturanyi	Stone loach	+		
Barbus prespensis	Prespa barbell	+		
Carassius gibelio	Prussian carp		+ (1970's)	
Chondrostoma prespensis	Prespa nase	+		
Cobitis meridionalis	Spined loach	+		
Ctenopharyngodon idella	Grass carp			+ (1980's)
Cyprinus carpio	Carp	+		
Gambusia holbrooki	Mosquito fish		+ (1960's)	
Hypophtalmichthys molotrix	Silver carp			+ (1980's)
Lepomis gibbosus	Pumpkinseed		+ (1995-1996)	
Oncorhynchus mykiss	Rainbow trout			+ (1970's)
Parabramis pekinensis	Bream			+ (1970's)
Pelasgus prespensis	Prespa minnow	+		
Phoxinus lumaireul	Minnow	+		
Pseudorasbora parva	Stone moroko		+ (1970's)	
Rhodeus amarus	Bitterling		+ (1990's)	
Rutilus prespensis	Prespa roach	+		
Salmo peristericus	Prespa trout	+		
Salmo letnica	Lake Ohrid trout			+ (1950's)
Silurus glanis	Catfish		+ (1980's)	
Squalius prespensis	Prespa chub	+		
Tinca tinca	Tench		+ (1980's)	

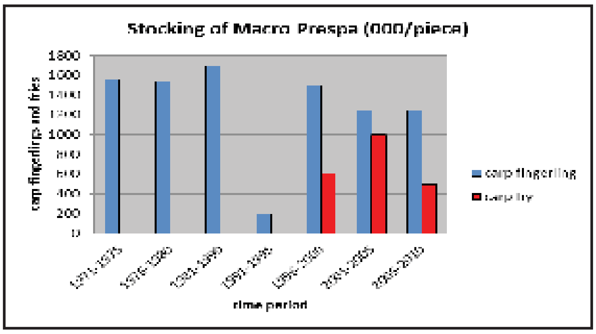
Out of determined 13 native, 8 fish species are endemics: Prespa spirlin, bleak, Prespa barbell, Prespa nase, Prespa minnow, Prespa roach, Prespa trout and Prespa chub.

At first sight, the proportion of endemism in the fish populations of the Prespa Lakes seems remarkable. It should be mentioned however, that, according to Crivelli et al. (1997), the taxonomic position of a number of taxa occurring in the Prespa Lakes remains doubtful. At present, only the barbell would appear to be undoubtedly endemic to both lakes, Micro and Macro Prespa (Dupont & Lambert, 1986; Economidis, 1989; Catsadorakis et al., 1996; Crivelli et al., 1996). Prespa barbell presents species with some economic importance in the Prespa watershed, with medium interest for fisheries (Kapedani & Hartmann 2009).

According to Economidis (1992) only two endemic species: Prespa barbell and Prespa trout are classified as "Endangered", which is an important criterion used for identification of priority species for conservation of animals in Prespa Region. Both species are listed as "Vulnerable" species on the IUCN Red List of Threatened Animals (Globally threatened species and Regional-European threatened species). Further, Prespa barbel is represented in Prespa Lakes in three countries (Macedonia, Albania, Greece) which is important for transboundary collaboration, unlike the Pelister stream trout that can be only found in the rivers in the Macedonian and Greek parts of Prespa basin (River Braychinska and its tributaries, Aghios Germanos stream, and others) (Crivelli et al., 2008).

The spawning season for most of the present fish species in the lakes is within the period April – June, with exception of the trout which spawns in the rivers from November – March. Another exception is the alien species pumpkinseed which spawns twice in the year – spring and autumn.

Stocking on Lake Prespa was performed only with autochthonous carp fingerlings starting from 1971 on both lakes Macro and Micro from the state owned hatchery in the village of Zvezda (Albania). From 1990 on, only Macro Prespa is stocked. On the following graphs stocked quantities for Macro and Micro Prespa from 1971 are presented. (Source: Transboundary Fish and Fisheries Management Plan. UNDP, 2012)



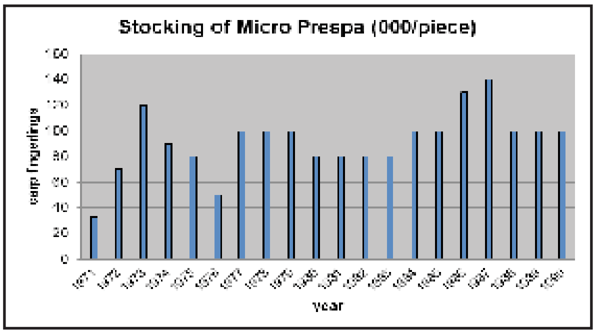


Figure 2. Macro Prespa stocked quantities of carp fingerlings and fries (1971-2010).

Figure 3. Micro Prespa stocked quantities of carp fingerlings (1971-1990).

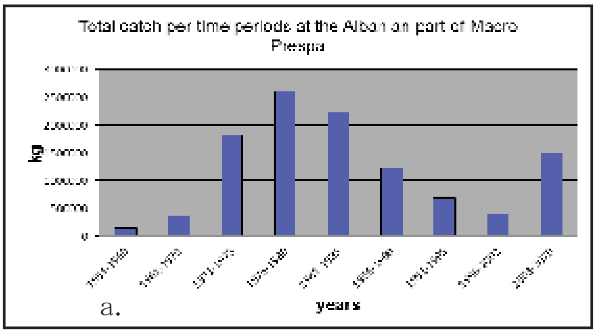
3.2. **Fisheries**

Fisheries have existed on the two Prespa Lakes since ancient time. It is recognized as artisan fishery. At present fishing is allowed on the Greek and Albanian parts of the Lakes, while on the Macedonian side there is still total ban. Two attempts as tendering procedures for concession of the fish were made (in 2012 and 2013), but none was successful.

3.2.1. **Albania**

There are 50 professional fishermen in Macro Prespa and 10 fishermen in Micro Prespa. However it should be emphasized that even the "professional" fishermen are also engaged in agriculture activities in their village during the time they are not in the lake.

Fishery statistics on Albanian part of the Macro Prespa Lake and Micro Prespa exist from 1954.



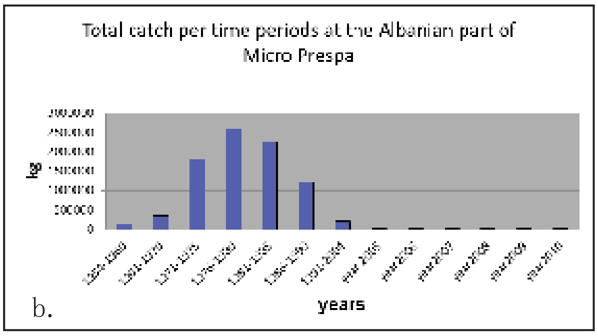


Figure 4. Total catch per time period (1954-2010) at the Albanian part of Macro Prespa (a) and Macro Prespa (b) (Source: Transboundary Fish and Fisheries Management Plan. UNDP, 2012)

Fish data collection is based on the Law 7908 dated 05/04/1995 as well as on the Regulation No. 1 dated 26/03/1997. The declaration of statistical data is one of the fundamental conditions for the renewal of fishing licenses. In the context of the further improvement of the data collection system on fisheries and of approaching Albanian legislation to that of the EU, preparatory work has begun on the improvement of the data collection system in the fisheries sector. It consists in the consideration given to the EC Regulations No. 1543/2000 dated June 29 2000 which determines the "Community structures for the collection and processing of necessary data in order to follow common policies in fisheries", as well as the EC Regulation No. 1639/2001 dated July 25, 2001 that establishes a "minimal program and a broad program for data collection in the fisheries sector and determines the ways to apply the Regulation (EC) 1543/2000". These regulations provide for the establishment of an efficient system of data collection as well as the development and funding of monitoring programs.

In reality hardly the system can be considered as reliable to several reasons; (i) large number of illegal fisherman's; (ii) no location for inspection of catch; (iii) the fleets are highly amortised; (iv) low awareness level and responsibility of fisherman's and (v) complicated marketing approaches; The following table is giving an overview of fish data for period 1954-2010.

Table 2. Composition of Fish Catch at the Albanian side of the Prespa Lakes 1954-2010 (Source: MoEFWA-Fishery)

Yewar	Carp	***Nase and other species	Bleak	Total Catch	Yield
	(%)	**(%)**	**(%)**	**(Kv)***	**(Kv/ha)**
1954-1960	20	13	67	1,500	0.287356322
1960-1970	13	5	82	3,700	0.708812261
1971-1975	3	6	91	18,072	3.462068966
1976-1980	0.5	4	95.5	25,989	4.978735632
1981-1985	0.5	3	96.5	22,415	4.294061303
1986-1990	4	5	91	12,177	2.332758621
1991-1995	5	8	87	6,933	1.32816092
1996-2000	6	2	92	6,200	1.187739464
2000-2010	4	2	94	6,300	1.206896552
*in the Original document the data are given as kv (*100= kg)					
**5220 ha water surface ***Nase and other species after 1991					

3.2.2. **Management and legislation**

There is no specific legislation for conservation of specific fish species, i.e. action plans). Other national laws, including fisheries related laws in Albania are giving a full set of actions that will secure species protection, such as:

Law no. 7664, dated 21.01.1993, concerning environmental protection

Law no. 7875, dated 23.11.1994, concerning protection of wild fauna and hunting

Law no. 7908, dated 5.4.1995 on fishing and aquatic life

Decision no. 80, dated 18. 02. 1999. Designation of Prespa as "National Park" and of Pogradeci as "Protected Landscape Area"

Law no. 8870, dated 21. 03. 2002. Amended by the law no. 7908 dated 5.04.1995, on "Fishing and Aquaculture"

Law No. 8763, dated 2. 04. 2001, concerning amendment of the law no. 7908, dated 5.04.1995, on "Fishing and Aquaculture"

Law no.8906, dated 6.6.2002, on protected areas

Law no.8934, dated 5.09.2002, on environmental protection

Law no. 9103, dated 10.7.2003, on protection of tranboundary lakes

Law no.9587, dated 20.07.2006, on biodiversity protection

Order no.262, dated 15.05.2006, approving the status of "Organization for Fish Management (OMP)"

Decision no. 146, dated 8. 5. 2007, on "Approving the Red List of Flora and Fauna".

Law no. 7, dated 15.01.2008, on fishery and aquaculture.

Law no. 87, dated 15.07.2008, on water.

The fishery sector and water resources regarding the management of the Albanian side of the lake is centralized. The management of all the lakes fisheries in Albanian territory is competence of the Ministry of Environment, Forestry and Water Administration, specifically of the Directorate of Fishery Policies. This

Ministry prepares the strategy for the lake fishery sector, the fisheries management plans and acts accordingly. For example this Ministry is responsible for issuing the fishing license for the lake and for the number of persons allowed to fish in the lake. There are in force also these legislative acts: Law No. 7908, date 5.4.1995 "**On Fishery and Aquaculture**", amended and Regulation No. 1 date 29.3.2005 **"For application of the legislation on fishery and aquaculture**" which determine the various regulating aspects for the Prespa Lake (MEFWA, 2009).

Regarding to the enforcement of the law and of the activities a fishing inspector responsible for the Korça district has the power and responsibility to control their enforcement. Other important aspect of this law is the set of the Fishing Inspectorate as the responsible and competent body in executing fishery laws, bylaws and regulations. The coordination of the inspection activities is responsibility of the Ministry, and the fishing Inspectorate is included as a division in the Directorate of Fishery Policy. The Inspector also reports on monthly basis in the Ministry for the status of the fishery activities in the lakes and the measures and penalties taken.

To enter in more details the specific management rules regarding the Prespa lake management present in the Albanian fishery legislation:

a) To achieve sustainable fish exploitation, the Directory of Fishery Policy has to prepare an administrative and development plan for the fishery and aquaculture sector.

b) To have a booking right in Professionals Fisherman Register, the requested person should, practice professional or seasonal fishing within a Fishery Management Organization.

c) In inland waters the license may be given to one or several boats, but the number have to be specified in the license.

d) The interruption of the fishing license is a competency of the fishing inspectors etc.

e) Catches by nets and hooks in Prespa have to be landed and traded first in centers approved by competent Veterinarian Authorities.

f) It is forbidden to fish, carry in board, to transit on the boat, purposed landing and trading with whatever mean and tool all fish species in Prespa lakes for a period of one month a year, issued by the Ministry*.

g) It is forbidden to fish carp from April 15 to May 15.

h) It is forbidden to fish and sell water organisms with dimension less then: *Alburnus* spp. 10 cm, *Chondrostoma* spp. 15 cm, *Rutilus* 12 cm, *Leuciscus* 15 cm, *Cyprinus carpio* 30 cm, *Carassius* spp. 15 cm etc.

i) It is prohibited to change water quality and the flow direction

j) It is prohibited to cut water vegetation without the approval of responsible bodies

k) It is forbidden to carry in boats or use nets by mesh size less than 66mm for carp in Prespa Lake.

3.2.3. **Macedonia**

The number of professional fishermen in Macro Prespa, with artisan organized cooperation till 2006, was 65 professional and 20 part-time active fishermen; usually in fruit farming as a main occupation.

Regarding the Lake Prespa mesotrophic character and relatively high fish production, annual yield for the period of commercial fishing was up to 363 tons and total annual catch differs from year to year and vary from 173 tons in the 60's to several tons in the latest period. The price of fish was relatively low through decades and in the period before the moratorium move from a minimum of 0.5 €/kg for bleak up to 3.5 €/kg for carp. Since 2007, there is no concession on Lake Prespa.

There is a fishery statistics for Prespa Lake regarding fish species dated from 1946.

Different species are predominant in the annual catch depending on the market demand and fishing gears used. In the period after Second World War trawls and seine nets were used, than in the beginning of 1960's purse seine net was introduced as a new fishing gear which results with higher percentage of bleak in the annual catch with 55 tons per year. Market demand for the same species in the 1990's derived with presence of more than 80% of the bleak in the commercial catch with 65 tons per year.

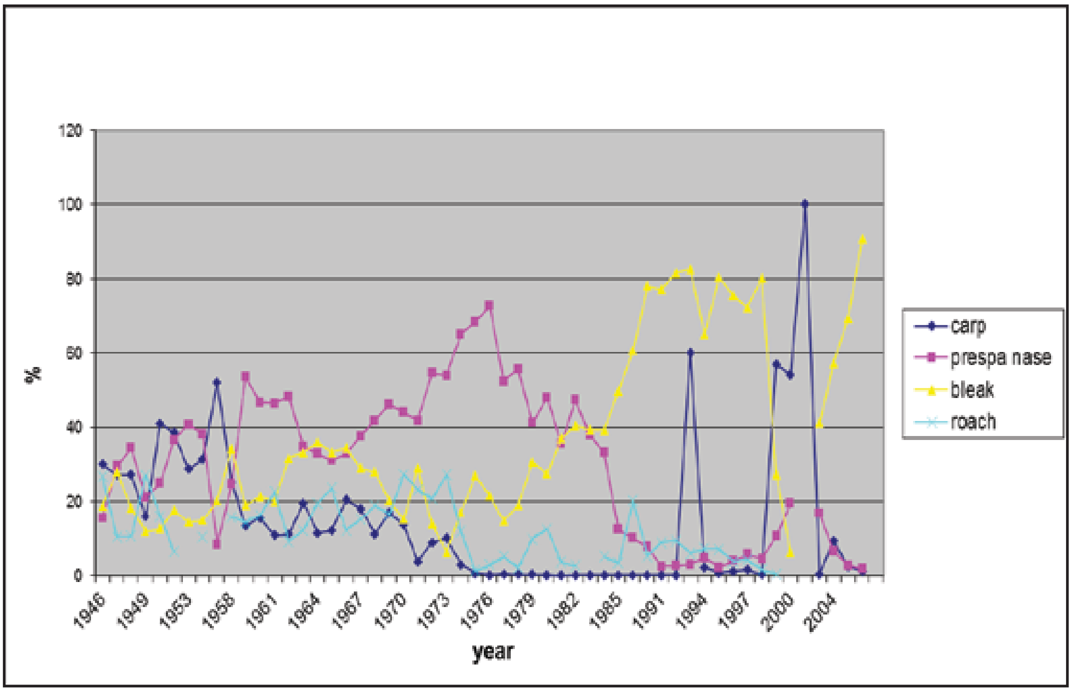


Figure 5. Presence of certain economically important fish species in the annual fish catch (1946-2006) at the Macedonian part of Macro Lake Prespa

3.2.4. **Management and legislation**

In 2007 the existing Law on Fishery (from 1993) has been replaced with the Law on Fishery and Aquaculture (LFA) Official gazette7/2008 date15.01.2008. This law has three amendments: one in 2010 Official gazette 67/2010 date 14.05.2010 and two in 2011: Official gazette 47/2011 date 08.04.2011 and 53/2011 date 14.04.2011.

The following documents are complimentary to the Law on Fishery and Aquaculture:

Law for the protection of Ohrid, Prespa and Dojran Lake Official gazette 45/1977 date 09.09.1977. This law has four amendments: one in 1980 Official gazette 08/1980, one in 1988 Official gazette 51/1988 and one in 1990, Official gazette 10/1990 and one in 1993, Official gazette 62/1993.

Law for nature protection Official gazette 67/2004 date 04.10.2004 This law has five amendments: one in 2006 Official gazette 14/2006, one in 2007 Official gazette 84/2007, one in 2010 Official gazette 35/2010,and two in 2011 Official gazette 47/2011 and Official gazette 148/2011.

Law for the environment Official gazette 53/2005 date 05.07.2005, This law has seven amendments: one in 2005 Official gazette 81/2005, one in 2007 Official gazette 24/2007, one in 2008 Official gazette 159/2008, one in 2009 Official gazette 83/2009, two in 2010 Official gazette 48/2010 and 124/2010 and one in 2011 Official gazette 51/2011.

Fishery Master Plan for Lake Prespa for the period 2011-2016. Official gazette 145/2011 and

Amendments of the Fishery Master Plan for Lake Prespa for the period 2011-2016. Official gazette 18/2013

Regulations:

Regulation on the form, content and the way of performing evidence of fish production as for the amount of the sold fish per species 2008

Regulation for performing the fish guarding service, the form and the content of the fish guardian legitimation, as the way of its issuing and withdrawing 2008

Regulation of the content of the Program for examining, the form and content of the certificate, as the cost for issuing certificate for commercial fishery 2008

Regulation on the form and the content of the evidence formulary in the fishing regions 2008

Regulation of the content of the Fishery Master Plan 2008

Regulation of the content of the annual plan for protection and exploitation of the fish and the content of the annual report of realization of the plan 2008

Regulation on the technical requirements for the lending sites 2008

Regulation on the quality, size and weight, as also the way of declaring the fish for traffic market 2008

Regulation on the way of marking of the boats and tagging and evidencing of the fishing gear 2008.

Regulation on the form and the content of the document for the origin of the fish and the way of its issuing and fulfilling 2010

Regulation on the way of issuing licenses for recreational fishing, the required documentation for issuing, the form and content of the evidence formulary, the way of evidencing and delivering the data 2010

Regulation on the form and the content of the legitimation for recreational fishing and the way of its issuing 2010

Regulation on the allowed fishing gears and equipment and their use for commercial and recreational fishing 2011

Regulation on the length of the fish under which they cannot be fished for commercial and recreational fishing 2011

Regulation on the quality, size and weight, as also the way of declaring the fish for traffic market 2013

Regulation for amendments of regulation on the allowed fishing gears and equipment and their use for commercial and recreational fishing 2013

Regulation for changes of the regulation on the length of the fish under which they cannot be fished for commercial and recreational fishing 2013

Within the Master Plan for the Macedonian Part of Lake Prespa (Official gazette of R. M. 145/211 and 18/2013 – issued by the Ministry of agriculture, forestry and water management and produced by PSI Hydrobiological Institute - Ohrid), protection of the fish and their habitats are on the first place. For these issues fishing bans per species are determined. At the same time Total Allowable Catch Quota (TACQ) per fish species was estimated on their minimum catchable length

(MCL). The number of required fishermen and fish guardians was stated also. Types of fishing and number of days and fishing gears per fisherman per species were determined. Commercial and recreational fisheries are allowed on the lake, while on the rivers only recreational one. Aquaculture activities within the lake are not allowed at all, while in the watershed only on autochthonous fish species of Prespa Basin. Total Allowable Catch Quota (TACQ) for commercial and recreational fishery on Lake Prespa is presented in the following tables.

Table 3. Commercial fishery TACQ

Common name	Latin title	Total allowable Catch quota per species in kg
carp	*Cyprinus carpio*	35.000
chub	*Squalius prespensis*	6.000
roach	*Rutilus prespensis*	15.000
bleak	*Alburnus belvica*	170.000
prussian carp	*Carassius gibelio*	unlimited
pumpkinseed	*Lepomis gibbosus*	unlimited
TOTAL		226.000

Table 4. Recreational fishery TACQ

Common name	Latin title	Total allowable Catch quota per species in kg
carp	*Cyprinus carpio*	4.000
chub	*Squalius prespensis*	3.000
roach	*Rutilus prespensis*	6.000
bleak	*Alburnus belvica*	18.000
prussian carp	*Carassius gibelio*	unlimited
pumpkinseed	*Lepomis gibbosus*	unlimited
TOTAL		31.000

Fishing gear for commercial fishing is limited to 15 bottom standing nets (one net has length of maximum 50 meters and height of maximum 5 meters with minimum mesh size of 45 mm) per fishermen for carp, 20 bottom standing nets (one net has length of maximum 45 meters and height of maximum 3 meters with minimum mesh size of 16 mm) per fishermen for bleak. For other commercial fish species as chub, roach and Prussian carp 15 bottom standing nets (one net has maximum length of 45 meters and height of 3 meters with minimum mesh size of 20 mm) per species per fishermen as well as trawling nets with maximum length of 500 meters and maximum height of 3 meters with mesh size of minimum 10 mm. For catfish fishing is allowed line with maximum 50 hooks per fishermen.

For recreational fishing - angling for all fish species is allowed fishing with two rods with one line with three hooks or three rods with one line and one hook.

3.2.5. **Comparative review of fishing/fishery rules in AL and MK**

Fishing ban season per species for Macedonian part of Prespa Lake is in 30 days in the spawning period which can differ from year to year, but has to be in the stated period in the following table.

Table 5. Fishing ban season by species and by countries

Common name	Latin name	ALBANIA		MACEDONIA	
carp	*Cyprinus carpio*	1st May	30th May	1st April	30th June
chub	Squalius prespensis	1st May	30th May	1st May	15th June
roach	*Rutilus prespensis*	1st May	30th May	1st May	30th June
bleak	*Alburnus belvica*	1st May	30th May	1st May	30th June

Table 6. Minimum allowed length for fishing of some commercial species

Common name	Latin name	ALBANIA	MACEDONIA
carp	*Cyprinus carpio*	30 cm	40 cm
chub	*Squalius prespensis*	15 cm	25 cm
roach	*Rutilus prespensis*	12 cm	15 cm
bleak	*Alburnus belvica*	10 cm	12 cm
prussian carp	*Carassius gibelio*	15 cm	unlimited
pumpkinseed	*Lepomis gibbosus*		unlimited

4. Transboundary sampling scheme performed

4.1. Methodology of sampling

The EN 14757 (multi-mesh gill nets - MMGN) standard was used to design the modified sampling scheme according to Lake Prespa specifications – large water body with different substrates (habitats). In order to satisfy statistical model we avoid periods of fish grouping (spawn and wintering shoaling).

Beside the MMGN in the planed protocol of CSBL project for Macedonian side of Lake Prespa, larval traps, fyke and beach seine nets were accepted to be used (ANNEX II), but due to procurement problems these fishing gears were not delivered.

4.1.1. Multi-mesh gillnetting

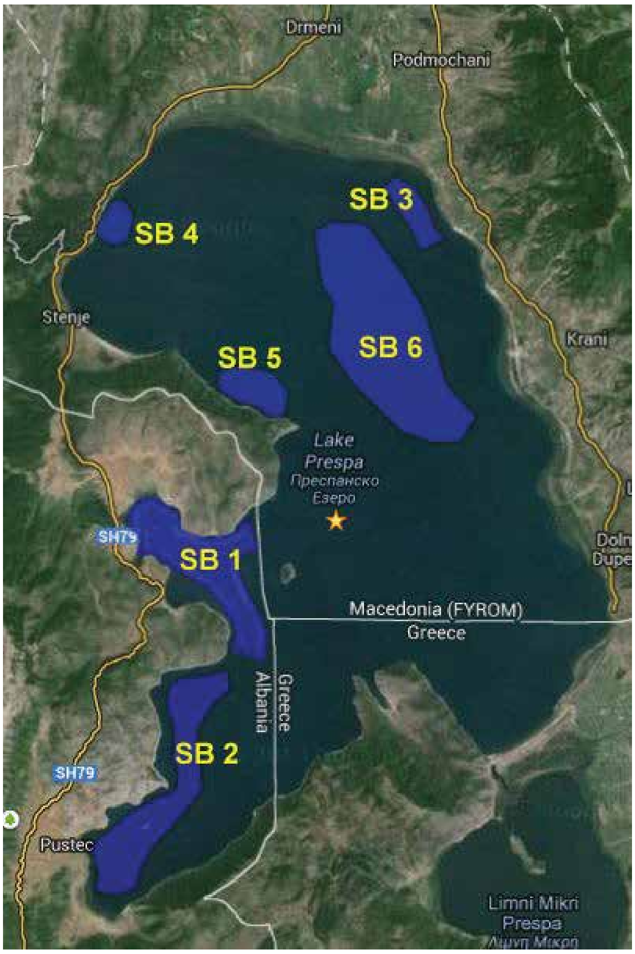


Figure 6. Lake Prespa sampling sub basins (SB1-SB6)

Fish population sampling was done according to the CEN 14757 standardized protocol, using benthic multi-mesh gillnets which are 30 m long and 1.5 m deep, composed of 12 panels with different mesh sizes ranging from 5 mm to 55 mm from knot to knot in the following order: 43mm, 19.5 mm, 6.25 mm, 10 mm, 55 mm, 8 mm, 12.5 mm, 24 mm, 15.5 mm, 5 mm, 35 mm and 29 mm.

Having in mind the lake's characteristics, bathymetry, habitat differentiation, and previous long term practise in experimental fishing, we divided the whole lake in 6 sub basins, SB 1-2 at Albanian side, and SB 3-6 at Macedonian side of the lake (Figure 6).

All SB's are different in habitat substrate, bathymetric configuration, wind exposures, and total ecological condition.

Five sub basins are littoral and one is pelagic sub basin.

Table 8. Lake Prespa sampling sub basins

Country	Sub basin (SB)	Locality	Position
Albania	**SB1**	Kallamas	littoral
	SB2	Liqenas	littoral
Macedonia	**SB3**	Asamati	littoral
	SB4	Otesevo	littoral
	SB5	Konjsko	littoral
	SB6	Central plate	pelagic

Lake Prespa sampling period was from 09 till 16. 10. 2013 at Macedonian part and from 16 till 28.10.2013 at Albanian part.

Nets were set before the dusk, stayed overnight and after the down were taken out (12 hours of sampling) to cover both highest activity circadian peaks. Nets were processed panel per panel, per species and measuring of the individuals was performed afterwards.

In both sampling sub sub-basins **SB1 (Kallamas)** and **SB2 (Liqenas)** which are two littoral sub basins, generally, a typical lake has three distinct zones of biological communities (limnetic, littoral and the benthic zone) linked to its physical structure. Rocky area covers coastal area at the south-western part of Macro Prespa where both sub – basins on Albanian part are belonging.

SB3 (Asamati) is a locality with the following habitat distribution:

a. From the lake shore up to 1.5 meter there is a fine muddy substrate and the whole area is covered with *Phragmites* (reed belt).

b. From the reed belt on up to 3 meters depth *Potamogeton* and *Myriophyllum* are present and substrate is muddy.

c. Third part of the littoral, from 3 to 6 meters depth is a muddy area.

This locality is under direct pressure from the tributary Golema Reka which is the main source of nutrient load from the agricultural area in its watershed.

SB4 (Otesevo) as the previous one has similar habitat distribution, but there is a big ecological difference since there is no tributary present, and also this part of the lake is not part of agricultural area of Prespa Lake watershed.

SB5 (Konjsko) is a locality with this habitat description:

1.From the lake shore up to 2.5 meters the substrate is consisted of rocks and gravel and vegetation present in this area is *Phragmites* and *Myriophyllum*.

2.In the zone from 2.5 meters up to 4 meters depth substrate is rocky with gravel and no vegetation.

3.From 4 meter till 12 m depth substrate is sandy.

The **SB6 (Central plate)** is a substitution of previously planned sub basin for multi-mesh pelagic nets sampling, and at this sub basin sampling was performed at deeper strata with benthic multi-mesh nets.

The planed sampling point for pelagic nets sampling – the deepest locality at Macro Prespa Lake (Kazan) with maximum depth of 34 m, was tested with different gill nets with one mesh size, but all nets were full with macrophyte vegetation (*Najas* sp.) and no fish catch.

4.1.1.1. **Albania**

The selection of sampling sites in the Albanian side of Lake Prespa and development of the sampling schedule are completed prior to the commencement of field work following the European standard (CEN, 2004 and CEN, 2005b).

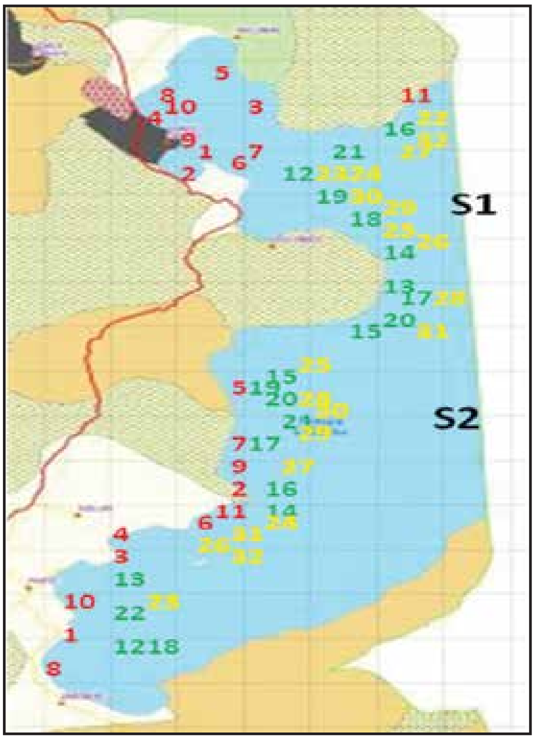


Figure 6. The positioning of benthic nets in two sub basins at the Prespa Lake. The red numbers – strata 0-3 m; green 3-6 and yellow 6-12 m.

Using the Lake Prespa surface area and maximum depth data we determined the total netting effort required according to the standard (CEN, 2004). The lake itself has been divided in two sub basins (respectively sub basin 1 and sub basin 2). Further to that the work was focused on Prespa lake bathymetric map (Topographic map 1:25 000 with grids over the map 250X250m). The grids were numbered starting from the first strata 0-3m, 3-6m and finally 6-12m. With this division over the map were produced a number of distinct, non-overlapping sampling units. The sampling sites are selected using a random numbers table.

The total number of 250m x 250m grids, including whole grids and portions of grids, which overlay the entire lakes surface has been assigned with their own unique number.

Selection of the sample sites was done using random number grids over the map begin selecting from the numbered grids with respective written letters and distributed over the map.

Following the depth strata (from bathymetric map 0-3m, 3-6m and 6-12m) and guidance (CEN, 2004 and CEN, 2005b) were set the number of nets in total 32 per sub basin 1 and 32 for sub basin 2.

The set of gillnets was organized in evening, and along with that the water temperature, transparency and coordinates are measured (Annex II).

The benthic multi-mesh gillnets were 30 m long and 1.5 m high, and composed of 12 different panels with mesh sizes ranging between 5 and 55 mm knot to knot in a geometric row.

The captured fish were identified to species level, counted and weighed in grams.

Total number of fish caught at two sub-basins SB1 and SB2 with this MMGN sampling campaign

is 15921.

Table 9. Summary table of multimesh gillnets sampling at two sub basins: Kallamas (SB1) and Liqenas (SB2)

WATER BODY	SUB BASIN Sampling date	No. of nets per sub basin	Maximum ind./net	Minimum ind./net	Nets / strata	
LAKE PRESPA	Kallamas (SB1) (16-21.10.2013)	32	1552	76	0-3 m	11
					3-6 m	11
					6-12 m	10
	Liqenas (SB2) (23-28. 10.2013)	32	1431	79	0-3 m	11
					3-6 m	11
					6-12 m	10

Figure 7. Pelagic nets testing

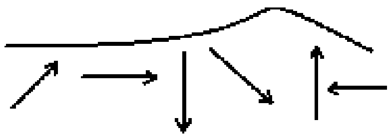


Figure 8. Random sampling

4.1.1.2. **Macedonia**

Sampling procedure was based on stratified random sampling. Depending on weather conditions, 8-10 benthic multi-mesh gillnets per night were set before dusk and were taken out after the down. Only testing of the pelagic nets was performed, but due to the high buoyance of those nets, for this sampling period they were not used. All pelagic nets are modified after the sampling campaign and can be used for future samplings.

For MMGN sampling at Macedonian part of the lake 10 of them were used with mesh size panels as described in the standard, but before usage previously we labelled each net with white cotton fabric with the number of the net from 1-10 and also each mesh panel with its dimensions for easier determination of the fish caught.

Figure 9. Lake Prespa fish sampling (MK)

GPS coordinates for each net, net setting depth, setting position to the shore, Air and Water temperature, pH, O_2, transparency – Sechi disk depth and weather conditions were registered (Annex II). All nets per strata following the randomization scheme were set in different directions related to the shoreline. So, for example in one particular strata, some nets were set from the shore starting with the panel of 43 mm, some nets ending closer to the shore with the panel of 29 mm in some cases perpendicular to the shore in some cases in an angle of 45° or 60°, in other cases parallel with the shore.

The Lake Prespa Station Monitoring Boat was used for setting and lifting the nets (64 in total) in sub basins on Macedonian side of Lake Prespa.

Table 10. Summary table of multimesh gillnets sampling at three sub basins: Asamati (SB3), Otesevo (SB4), Konjsko (SB5) and Central plate (SB6)

WATER BODY	SUB BASIN Sampling date	No. of nets per sub basin	Maximum ind./net	Minimum ind./net	Nets /strata	
LAKE PRESPA	**ASAMATI (SB3)** **(11.10.2013/16.10.2013)**	15	150	22	0-3 m	5
					3-6 m	5
					6-12 m	5
	OTESEVO (SB4) **(10.10.2013/11.10.2013)**	12	1606	14	0-3 m	4
					3-6 m	4
					6-12 m	4
	KONJSKO (SB5) **(12.10.2013/15.10.2013)**	12	548	72	0-3 m	5
					3-6 m	3
					6-12 m	4
	CENTRAL PLATEAU **(13.10.2013/14.10.2013/15.10.2013)**	25	70	1	14-16 m	25
	Pelagic net	1	181		0-6 m	1

Figure 10. Catch processing

Considering how many individuals per species per panel were caught, in those sub basins where there were not more than 50 individuals, all caught specimens were measured. In cases where several hundreds of one species were caught per panel, 50 individuals were measured by length and weight and the total weight and number of individuals of the rest was recorded. Weight was measured on a portable balance with accuracy of 0.1 g. Standard and total length were measured to the closest mm and for data processing just total length was used and averaged to the closest cm. In total 5144 individuals were caught during the Lake Prespa multi-mesh gillnets sampling campaign for this project on Macedonian part of the lake.

4.2 Data analysis and management

4.2.1 Albania

Captured fish were identified to species level, counted and weighed in grams. A comprehensive fact sheet of each sampling location has been filled following the previous agreements during the workshops CSBL Project with counting measuring weighting all the fishes per each net. The excel files were later on elaborated following agreement from the reporter leader (in case of Prespa HBI Ohrid) and in line with outline and guideline provided by the CSBL.

CPUE expressed in biomass of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m2(BPUE) and individuals of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m^2(NPUE).

A literature review considered past studies with analytical approaches to the assessment of freshwater fish communities, particularly those relevant to the requirements of CSBL and the WFD

4.2.2. Macedonia

All data were recorded during the field measuring at hard copy documents (Annex data - copy of the Lake Prespa data sheets). Data from these data sheets were entered into the Excel files for separate sub basin, per net, per panel, per species (length and weight). Further on analysis of the data was performed for fish species composition per sub basin, fish species abundance per depth strata in the sub basin, CPUE expressed in biomass of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m2(BPUE) and individuals of species per depth strata (g/m^2) per net surface 1,5m x 30m = 45m^2(NPUE).

Species evenness and richness between sub basins were obtained from data and also species length frequency for all species caught.

All processed and analysed data are archived as data sets that can be used for future data base development.

4.3. **Results**

To adopt the standard EN 14757 from one side and to fulfil the same for large water body as Lake Prespa is, random sampling with MMGN was performed on six different (by habitats, wind exposure, water currents, ecological conditions, addition nutrient load and exposure to harmful substances) sub-basins. In total 128 nets were set at both sampling parts of the Lake, 64 per country.

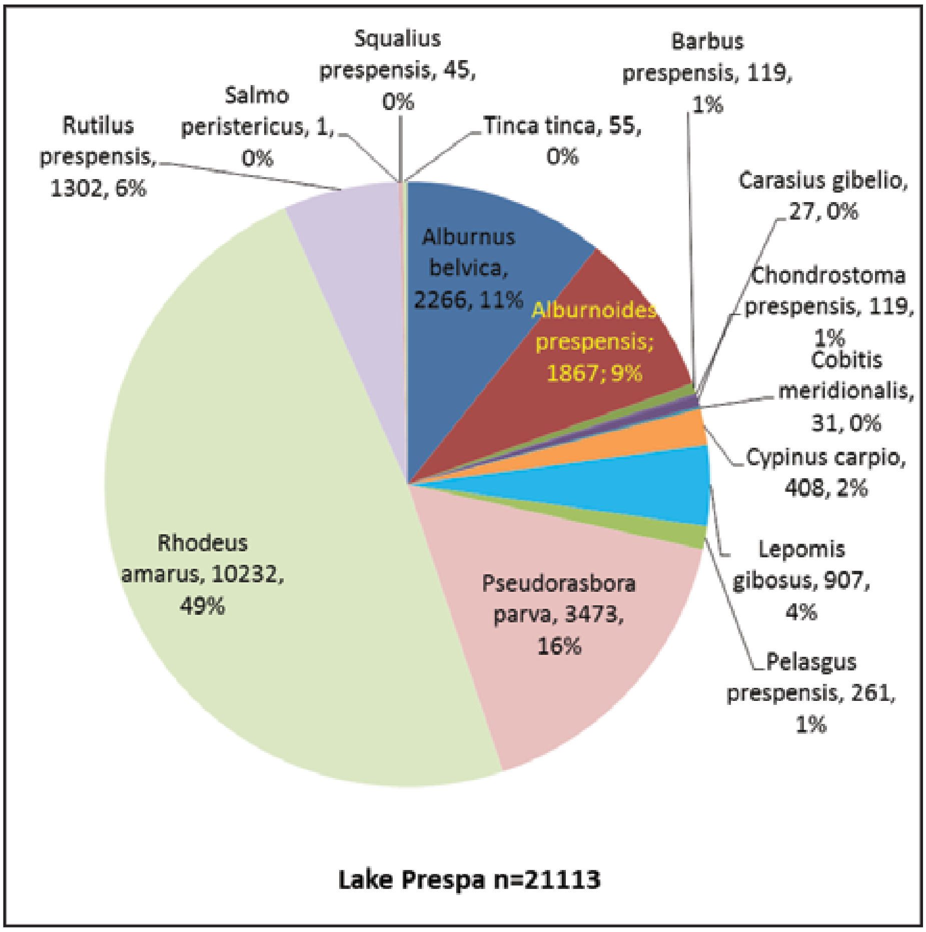
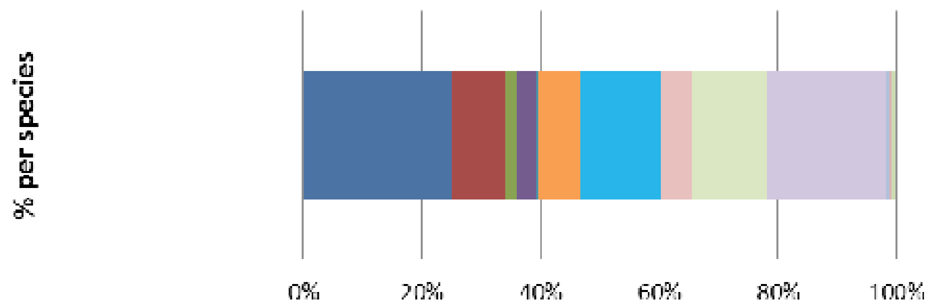


Figure 11. Relative and absolute fish species composition represented in the total catch of Lake Prespa (SB1-SB6) during the sampling campaign in October 2013

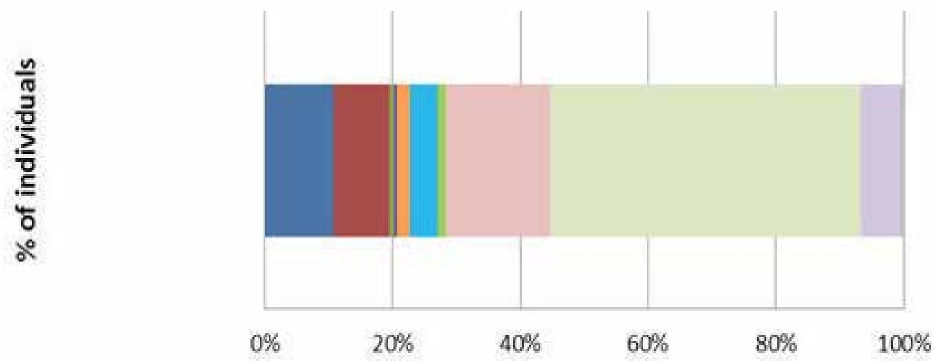
This figure in fact represents the actual (instantaneous – ten days sampling campaign) fish assemblage in October 2013. In total, 15 fish species were recorded in the catch. These results which are showing dominance of the alien species (see Tab. 1) with more than 69% of the total catch can serve mainly about determining the ecological status of certain habitats and also knowledge of the alien species ecology in relation to manage or design certain strategies for their reduction. From other side, regarding fishery issues (catch quotas, minimum catchable size, types of gears or even simple restricted fish stock assessment) the obtained data are not usable.



	g/m²
Alburnus belvica	223.38
Alburnoides prespensis	79.89
Barbus prespensis	15.37
Chondrostoma prespensis	31.54
Cobitis meridionalis	0.75
Cyprinus carpio	65.24
Lepomis gibbosus	119.97
Pelasgus prespensis	2.17
Pseudorasbora parva	42.45
Rhodeus amarus	114.65
Rutilus prespensis	176.17
Salmo peristericus	0.36
Squalius prespensis	8.60
Tinca tinca	2.68
Carassius gibelio	6.00

Figure 12. CPUE expressed in biomass (g/m²) in percentage per species of total catch per sub-basin (BPUE) of Lake Prespa during the sampling campaign in October 2013

Taking Lake Prespa as one water body (in this case Albanian and Macedonian part), regarding the biomass of fish species, two native (bleak and roach) and two alien species (pumpkinseed and bitterling) are predominant. On the other side, regarding the NPUE stated alien species are predominant.



Species	ind./m²
Alburnus belvica	0.39
Alburnoides prespensis	0.32
Barbus prespensis	0.02
Chondrostoma prespensis	0.02
Cobitis meridionalis	0.01
Cyprinus carpio	0.07
Lepomis gibbosus	0.16
Pelasgus prespensis	0.05
Pseudorasbora parva	0.60
Rhodeus amarus	1.78
Rutilus prespensis	0.23
Salmo peristericus	0.00
Squalius prespensis	0.01
Tinca tinca	0.01
Carassius gibelio	0.00

ind./m^2

Figure 13. CPUE expressed in number of individuals/m^2 in percentage per species of total catch per sub-basin (NPUE) of Lake Prespa during the sampling campaign in October 2013

4.3.1. Albania

4.3.1.1. Multi-mesh gillnetting

4.3.1.1.1. Abundance and fish assemblage

During fish sampling campaign on two sub-basins SB1 (Kallamas) and SB2 (Liqenas) following 15 fish species were recorded: Prespa bleak, Prespa spirlin, Prespa barbell, Prespa nase, spined loach, Prespa minnow, Prespa roach, Prespa trout, Prespa chub, carp, Prussian carp, pumpkinseed, bitterling, stone moroko and tench.

Note: a matured 71 kg individual of catfish has been caught out of our campaign sampling on 11th November in Kallamas.

In both sub basins, SB1 and SB2 bitterling is representing a dominant species in terms of number of individuals particularly at the depth strata 0-3 and 3-6 meters with 51% of total number of individuals.

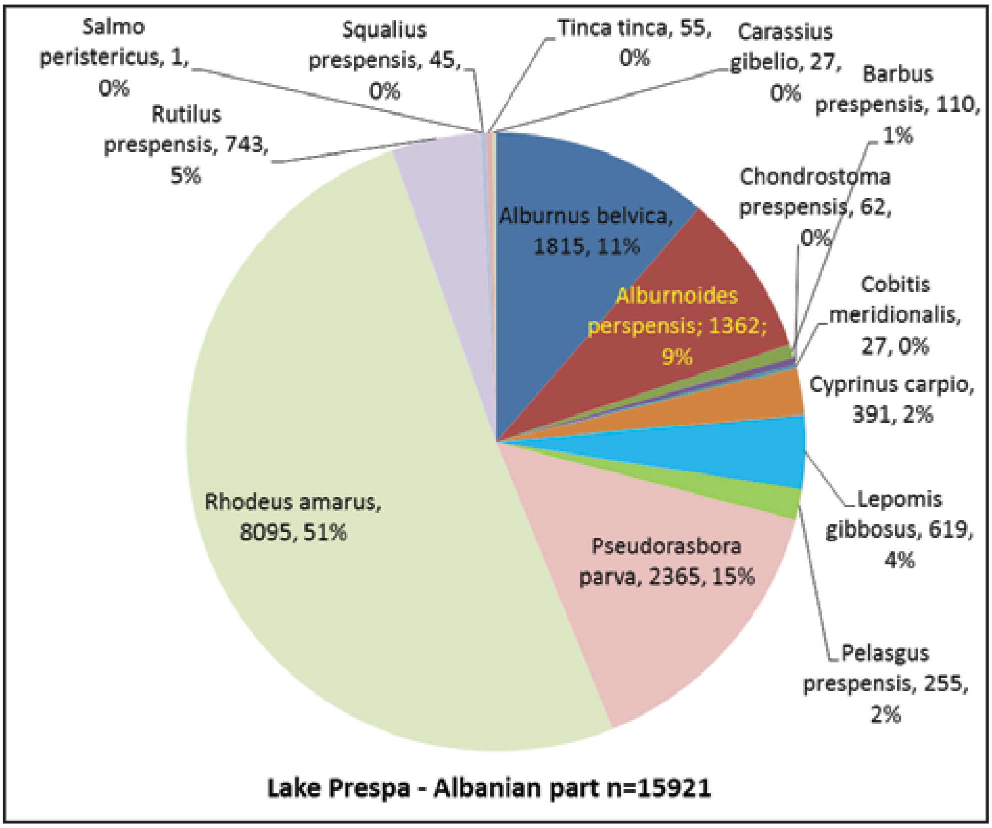
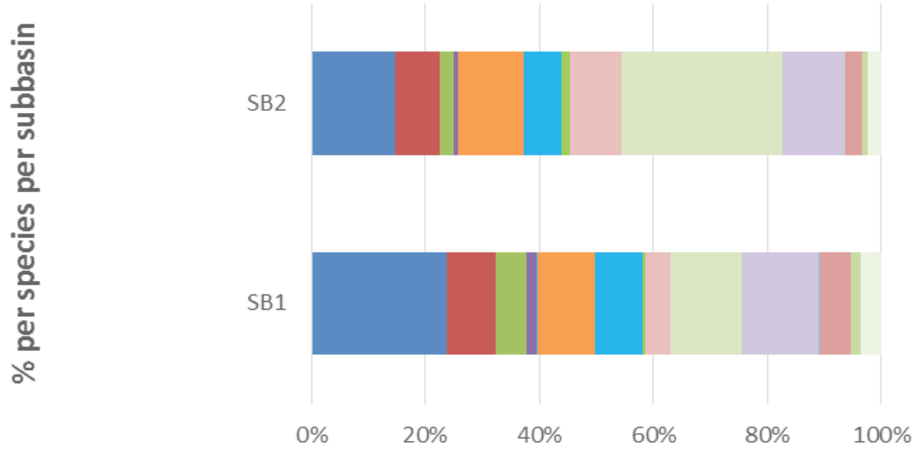


Figure 14. Relative and absolute fish species composition represented in the total catch at the Albanian Part of Lake Prespa during the sampling campaign in October 2013

The second most present species in terms of numbers from the alien's one is stone moroko that for entire sampled area represents 15%.

4.3.1.1.2. **CPUE**

Differences between two sampled sub-basins are only present in the biomass/m^2 where dominant species for SB1, bleak and bitterling are presented with almost the same biomass as bitterling in the SB2. As for the third dominant species native roach, both SB are showing similar values.

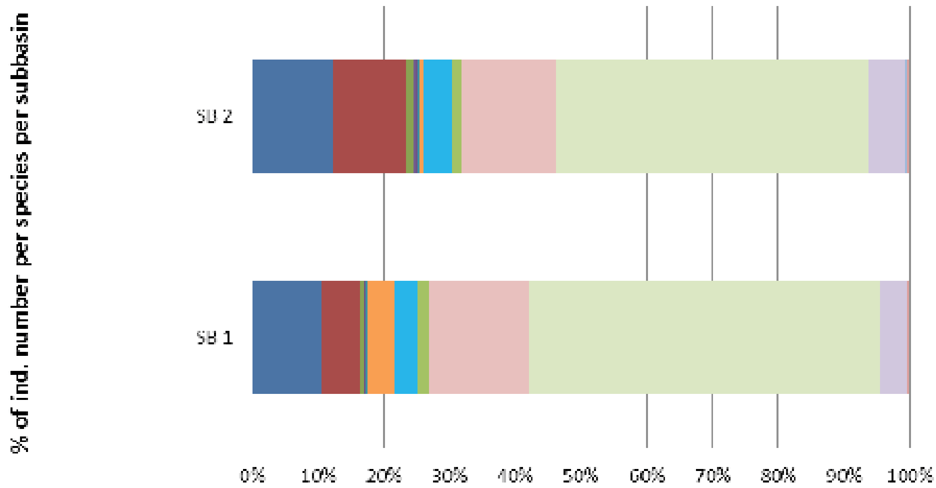


	SB1	SB2
Alburnus belvica	29.66	9.33
Alburnoides prespensis	10.94	4.98
Barbus prespensis	6.53	1.55
Chondrostoma prespensis	2.28	0.39
Cobitis meridionalis	0.24	0.10
Cyprinus carpio	12.76	7.25
Lepomis gibbosus	10.22	4.22
Pelasgus prespensis	0.82	0.88
Pseudosabora parva	5.26	5.78
Rhodeus amarus	15.80	17.95
Rutilus prespensis	16.96	7.04
Salmo peristericus	0.36	0.00
Squalius prespensis	6.71	1.89
Tinca tinca	2.11	0.57
Carassius gibelio	4.48	1.51

g/m^2

Figure 15. CPUE expressed in biomass (g/m^2) in percentage per species of total catch per sub-basin (BPUE) at Albanian Part of Lake Prespa during the sampling campaign in October 2013

Regarding the following figure which is expressing the catch per unit effort in numbers of individuals per square meter of net, the actual recorded diferences are only in the values of Prespa spirlin and carp.



	SB 1	SB 2
Alburnus belvica	0.61	0.65
Alburnoides perspensis	0.34	0.60
Barbus prespensis	0.03	0.05
Carassius gibelio	0.01	0.01
Chondrostoma prespensis	0.02	0.02
Cobitis meridionalis	0.01	0.01
Cyprinus carpio	0.24	0.03
Lepomis gibbosus	0.21	0.22
Pelasgus prespensis	0.09	0.09
Pseudorasbora parva	0.89	0.75
Rhodeus amarus	3.10	2.57
Rutilus prespensis	0.23	0.28
Salmo peristericus	0.00	0.00
Squalius prespensis	0.01	0.07
Tinca tinca	0.02	0.02

ind./m^2

Figure 16. CPUE expressed in number of individuals/m^2 in percentage per species of total catch per sub-basin (NPUE) at Albanian Part of Lake Prespa during the sampling campaign in October 2013

4.3.1.1.3. **SB1 – Kallamas**

According the EN 14757 standard, 32 multi-mesh gillnets were set randomly at SB1 in three strata: 0-3 meters (11 gillnets), in the strata 3-6 meters (11 gillnets) and in strata 6-12 meters (10 gillnets). As a result of this sampling campaign at SB1 bitterling is representing a dominant species in terms of number of individuals particularly at the depth strata 0-3 and 3-6 meters with 48%. Second dominant species is another introduced species, stone moroko with 14% followed by two native species bleak and spirlin with 12 and 11% respectively.

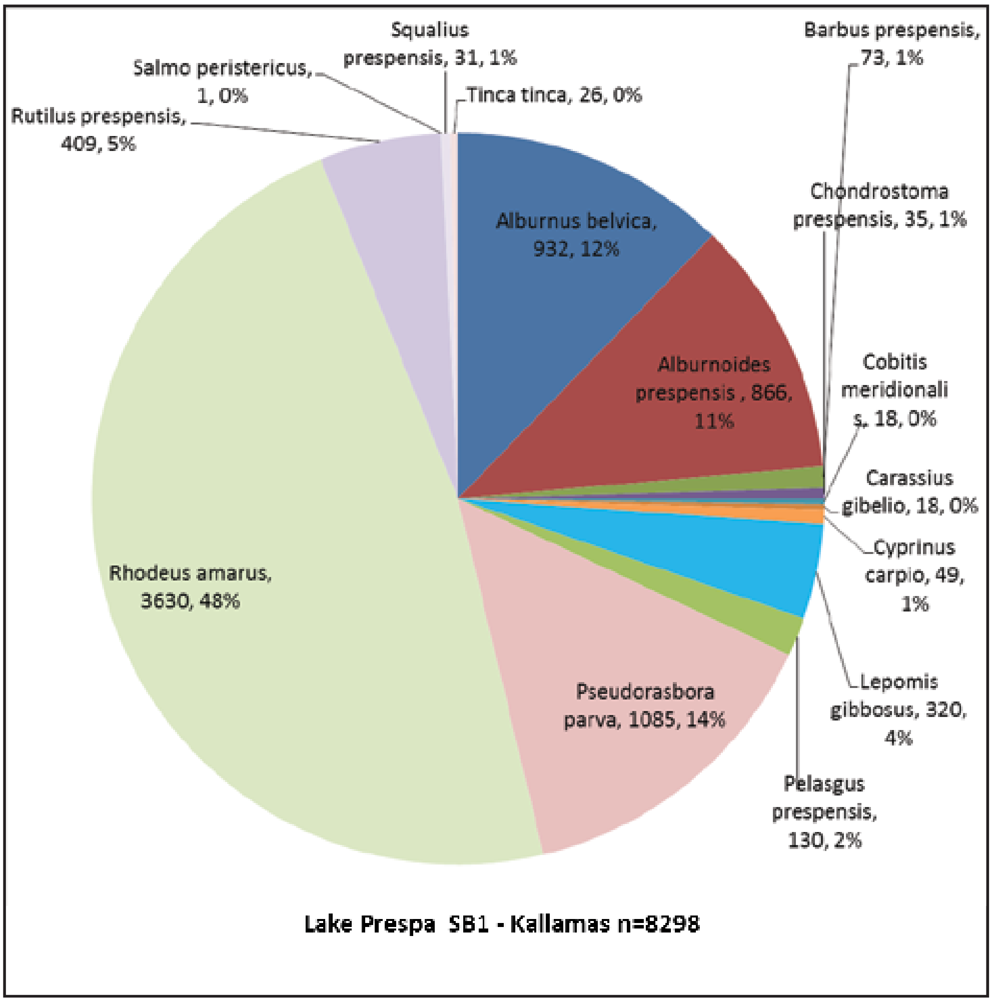
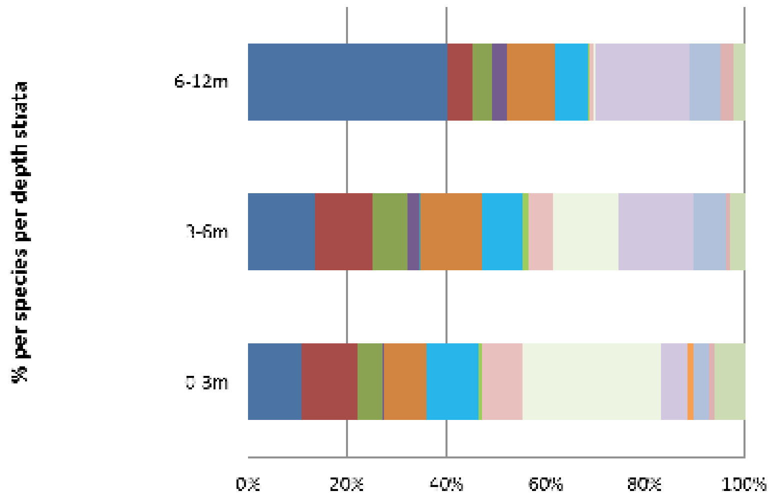


Figure 17. Relative fish species composition in total catch at SB1 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 54 individuals)

There is a clear difference in species dominance ones sampling is progressing from 0-3 meters to 6-12 meters, where at the deepest part the native endemic species bleak and roach, are composing about 63% of entire CPUE in biomass per square meter of net.

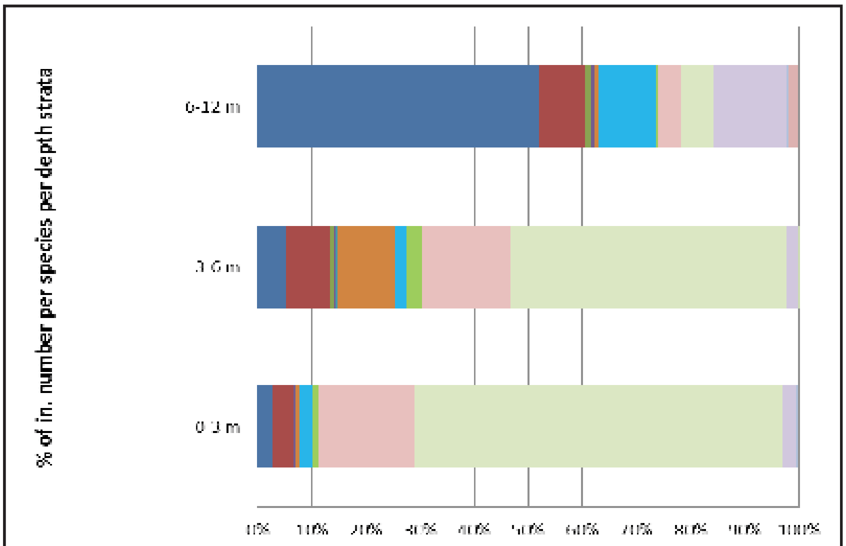


	0-3m	3-6m	6-12m
Alburnus belvica	4.22	4.55	20.89
Alburnoides prespensis	4.43	3.97	2.54
Barbus prespensis	1.93	2.39	2.21
Chondrostoma prespensis	0.09	0.77	1.43
Cobitis meridionalis	0.04	0.19	0.00
Cyprinus carpio	3.40	4.20	5.16
Lepomis gibbosus	3.93	2.80	3.48
Pelasgus prespensis	0.34	0.31	0.17
Pseudosabora parva	3.21	1.80	0.24
Rhodeus amarus	11.00	4.40	0.40
Rutilus prespensis	2.06	5.12	9.78
Salmo peristericus	0.36	0.00	0.00
Squalius prespensis	1.33	2.24	3.15
Tinca tinca	0.39	0.28	1.44
Carassius gibelio	2.35	0.99	1.14

g/m²

Figure 18. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 1 locality

As it has been stated above both species bitterling and stone moroko are the most abundant species in terms of number of individuals, while other alien species like pumpkinseed tench and Prussian carp are les abundant.

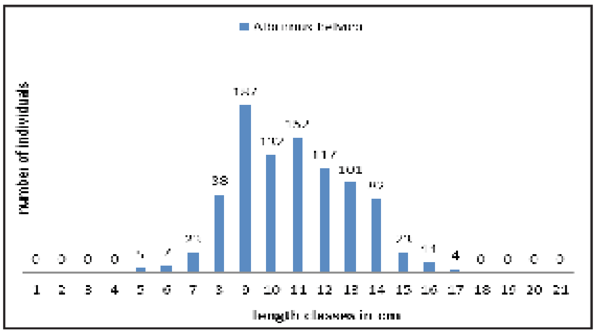


	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.23	0.31	1.38
Alburnoides prespensis	0.35	0.45	0.22
Barbus prespensis	0.02	0.03	0.03
Chondrostoma prespensis	0.01	0.03	0.07
Cobitis meridionalis	0.00	0.01	0.00
Cyprinus carpio	0.06	0.60	0.02
Lepomis gibosus	0.23	0.12	0.28
Pelasgus prespensis	0.08	0.17	0.00
Pseudorasbora parva	1.57	0.92	0.11
Rhodeus amarus	6.00	2.88	0.16
Rutilus prespensis	0.24	0.10	0.36
Salmo peristericus	0.00	0.00	0.00
Squalius prespensis	0.02	0.00	0.01
Tinca tinca	0.01	0.01	0.04
Carasius gibelio	0.01	0.00	0.00

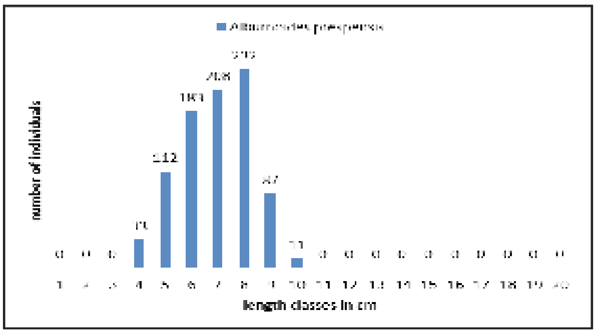
ind./m²

Figure 19. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 1 locality

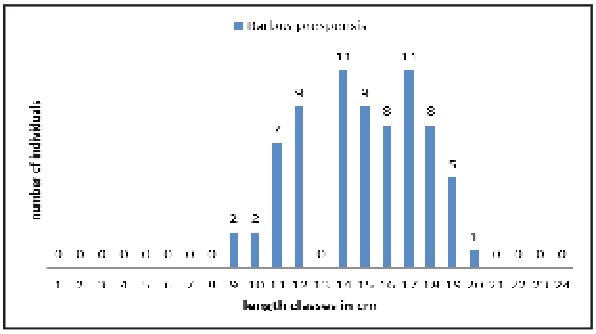
The length frequency distributions of the fish species caught during the survey at the SB 1 are presented in the following figure (a-j).



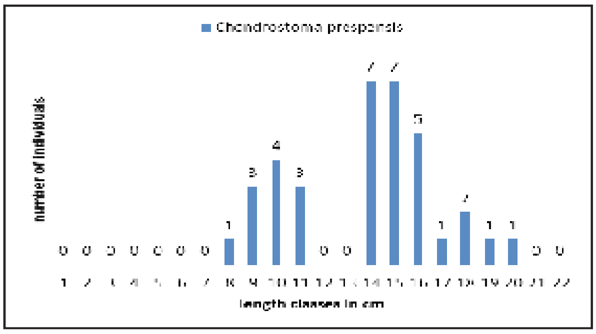
a)



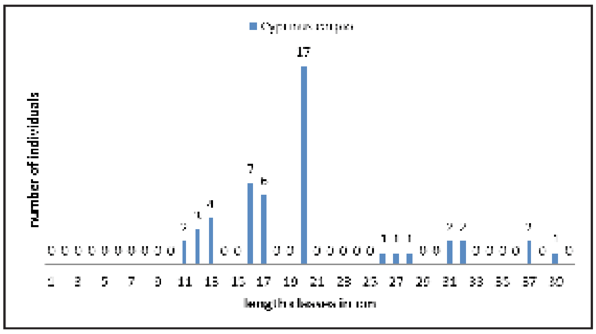
b)



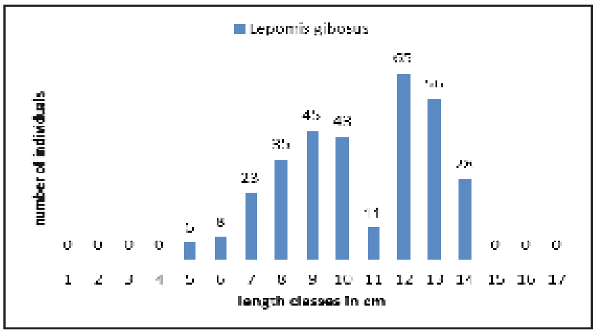
c)



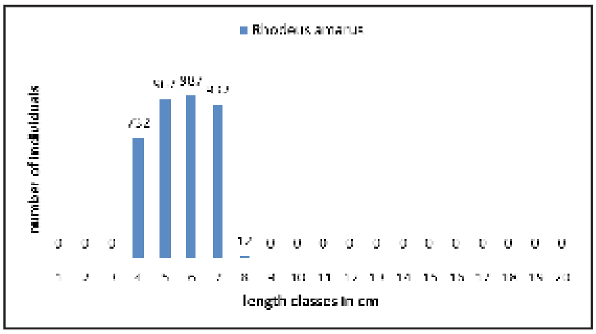
d)



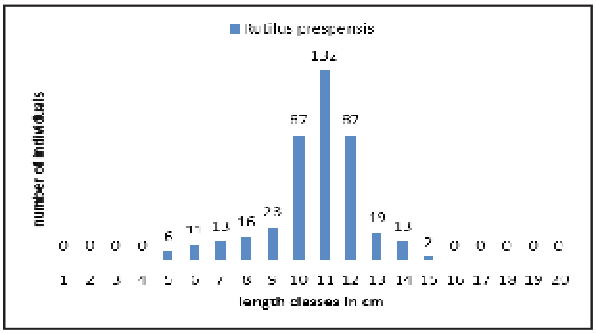
e)



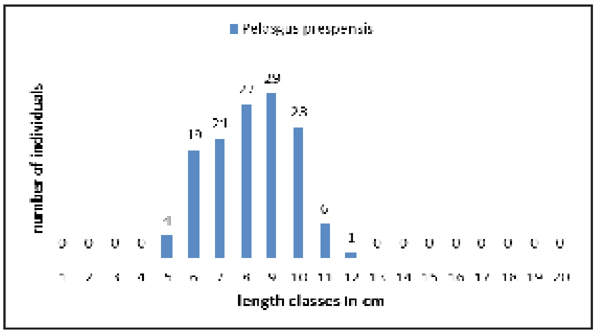
f)



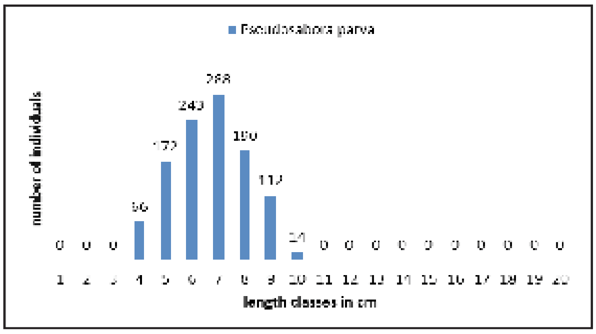
g)



h)



i)



j)

Figure 20. Length classes frequency per species for the SB 1 Kallamas (from a to j)

4.3.1.1.4. **SB2 - Liqenas**

Same numbers of nets with same distribution per strata were set at SB2 as were for SB1. Similar results were obtained for both sub-basins. As it has been stated above both alien species bitterling and stone moroko are the most abundant species in terms of number of individuals with 54 and 15% respectively, while other alien species like pumpkinseed, tench and Prussian carp are less abundant.

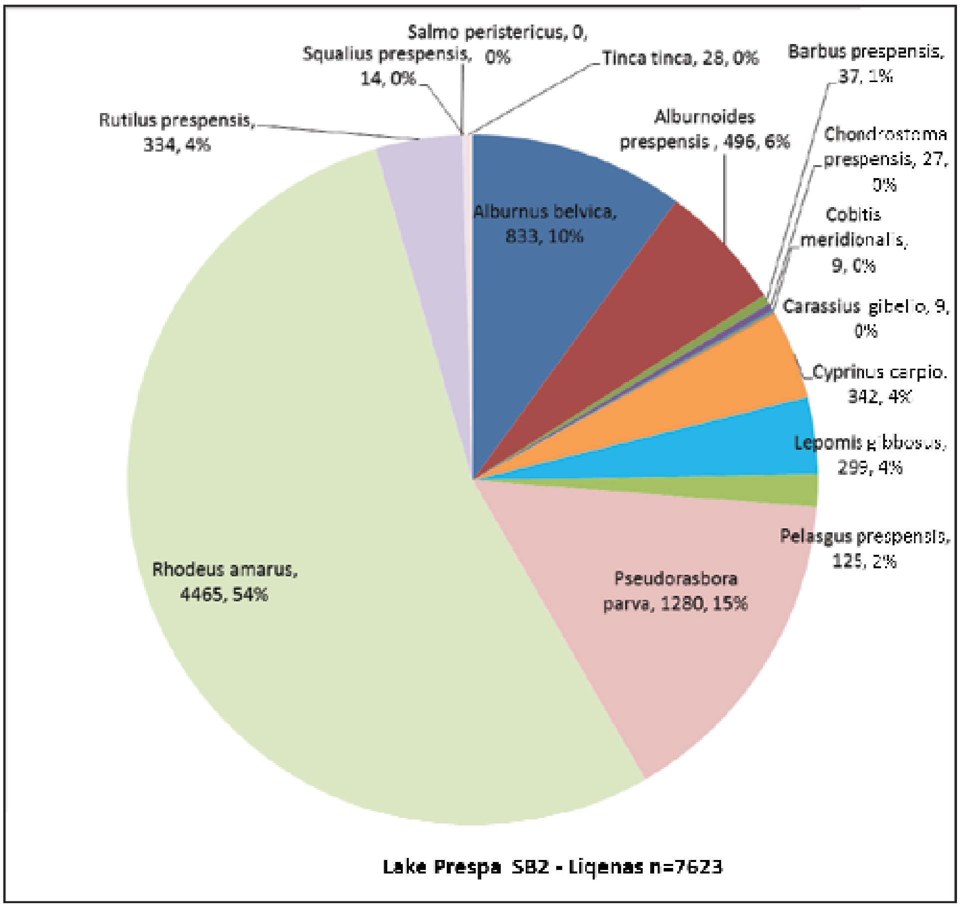
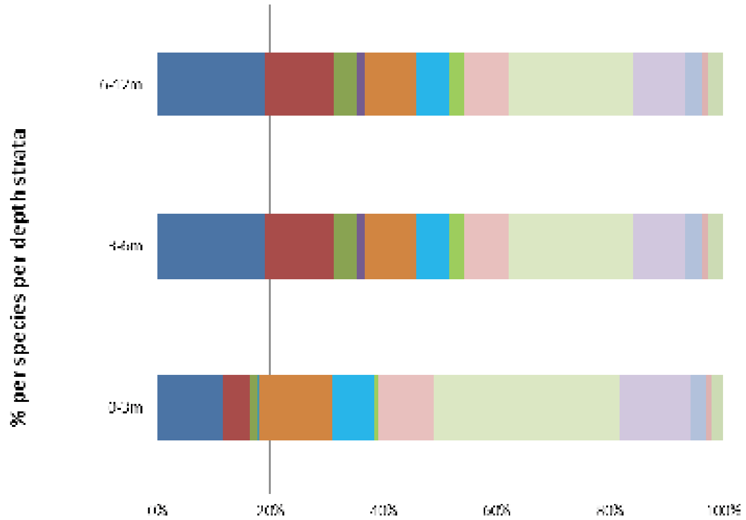


Figure 21. Relative fish species composition in total catch at SB2 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 28 individuals)



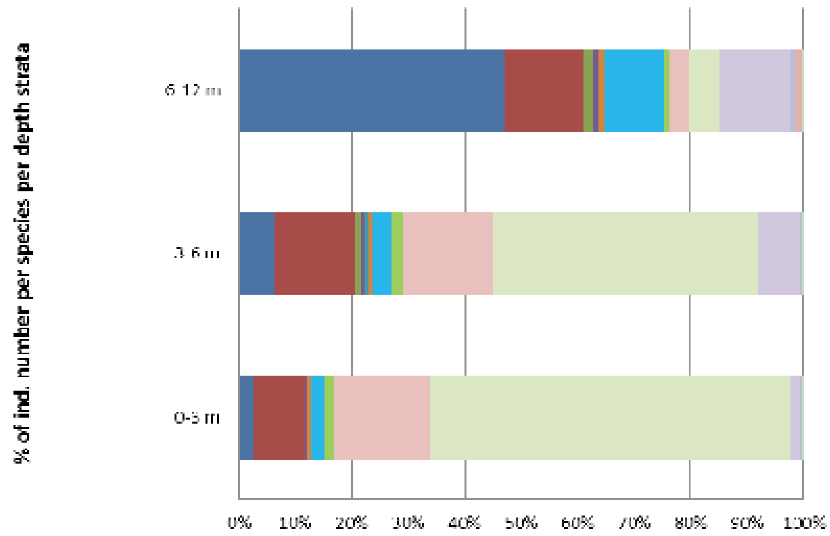
	0-3m	3-6m	6-12m
Alburnus belvica	4.26	4.06	0.11
Alburnoides prespensis	1.83	3.09	0.07
Barbus prespensis	0.47	1.06	0.02
Chondrostoma prespensis	0.07	0.32	0.01
Cobitis meridionalis	0.03	0.06	0.00
Cyprinus carpio	4.82	2.38	0.05
Lepomis gibbosus	2.74	1.45	0.03
Pelesgus prespensis	0.22	0.65	0.01
Pseudosabora parva	3.64	2.09	0.05
Rhodeus amarus	12.21	5.61	0.12
Rutilus prespensis	4.57	2.41	0.05
Salmo peristericus	0.00	0.00	0.00
Squalius prespensis	1.14	0.74	0.02
Tinca tinca	0.28	0.28	0.01
Carassius gibelio	0.79	0.71	0.02

g/m²

Figure 22. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 2 locality

Carp presence in terms of biomass per square meter of net for the entire depths varies from 16% at the depth strata 0-3 m, 12% at the strata 3-6 m and 10 % at the deepest sampled strata 6-12 m.

For the catch per unit effort in numbers of individuals per square meter of net (NPUE) there is a clear difference in species dominance ones sampling is progressing from 0-3 meters to 6-12 meters, where at the deepest part the native endemic species bleak and roach, are composing about 63% of entire catch for the depth strata. The bitterling is in all three depth strata the most dominant species.

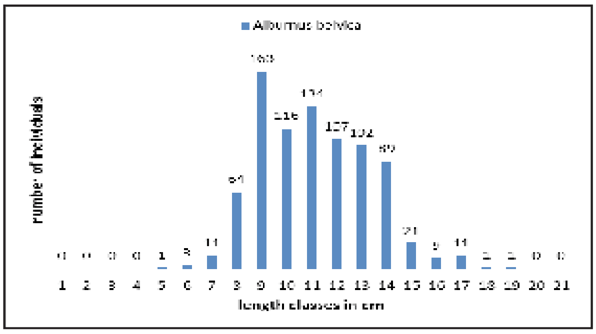


	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.21	0.30	1.51
Alburnoides perspensis	0.70	0.65	0.44
Barbus prespensis	0.04	0.06	0.05
Chondrostoma prespensis	0.00	0.03	0.04
Cobitis meridionalis	0.01	0.03	0.00
Cyprinus carpio	0.05	0.02	0.03
Lepomis gibosus	0.18	0.16	0.34
Pelasgus prespensis	0.14	0.09	0.03
Pseudorasbora parva	1.34	0.75	0.11
Rhodeus amarus	4.99	2.19	0.17
Rutilus prespensis	0.13	0.34	0.39
Salmo peristericus	0.00	0.00	0.00
Squalius prespensis	0.02	0.02	0.02
Tinca tinca	0.01	0.00	0.05
Carasius gibelio	0.02	0.01	0.01

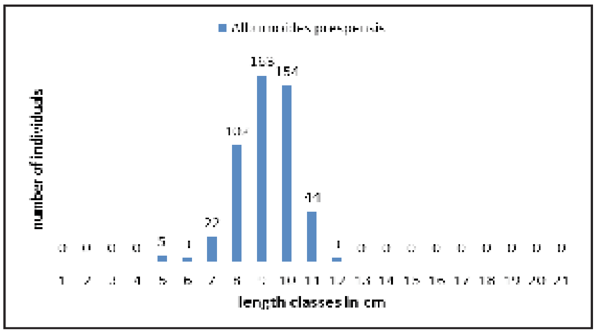
ind./m²

Figure 23. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 2 locality

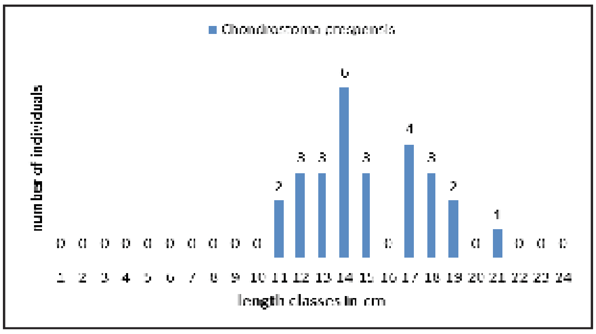
The length frequency distributions of the fish species caught during the survey at the SB 2 are presented in the following figure (a-j).



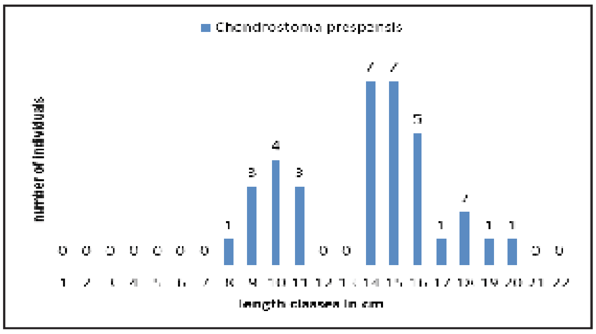
a)



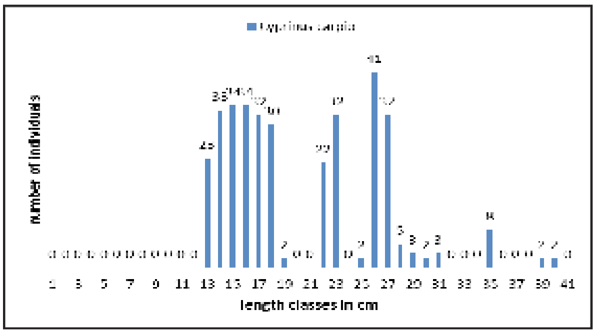
b)



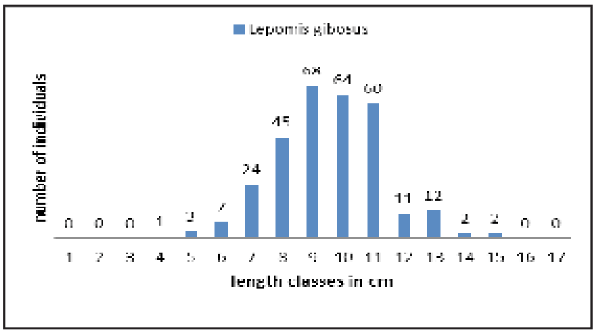
c)



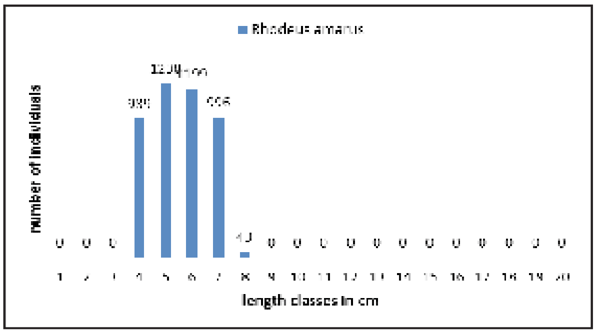
d)



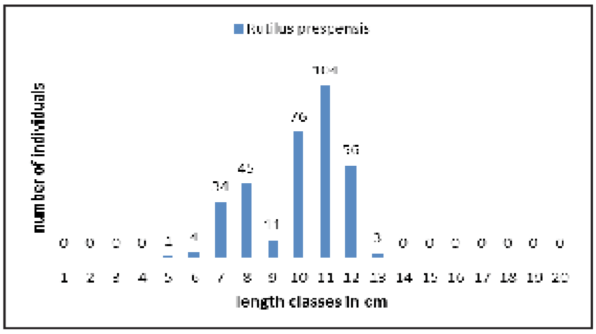
e)



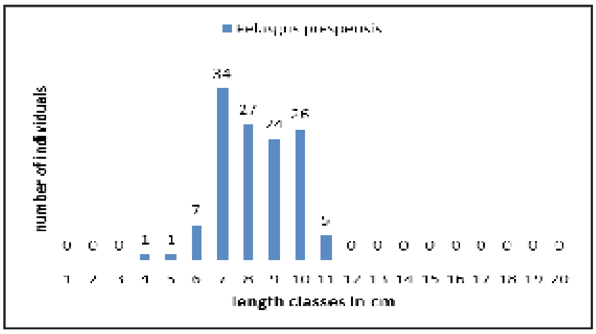
f)



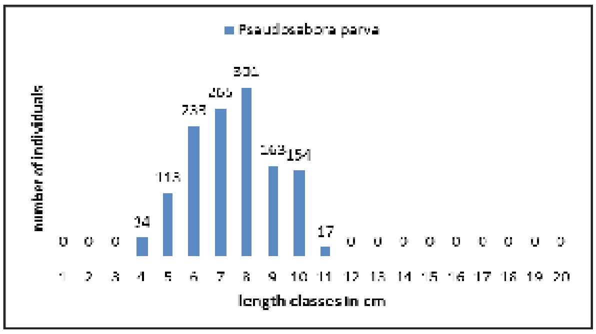
g)



h)



i)



j)

Figure 24. Length classes frequency per species for the SB 2 Liqenas (from a to j)

The bleak caught during this sampling campaigning ranged from 77 mm to 168 mm. For the barbell as it is shown at the Figure 24 (c) there are two clusters of size classes 97-172 mm and 188-210. Presence of matured individuals is a good indicator for the species recovery after the colaps recorded in the last decade.

4.3.2. **Macedonia**

4.3.2.1. **Multimesh gill netting**

The sampling campaign at the Macedonian part of Lake Prespa was performed at four sub basins Asamati, Otesevo, Konjsko and Central Plate. The first three were with more or less same surface area with exception of the one in the Central Plate.

4.3.2.1.1 **Abundance and fish assemblage**

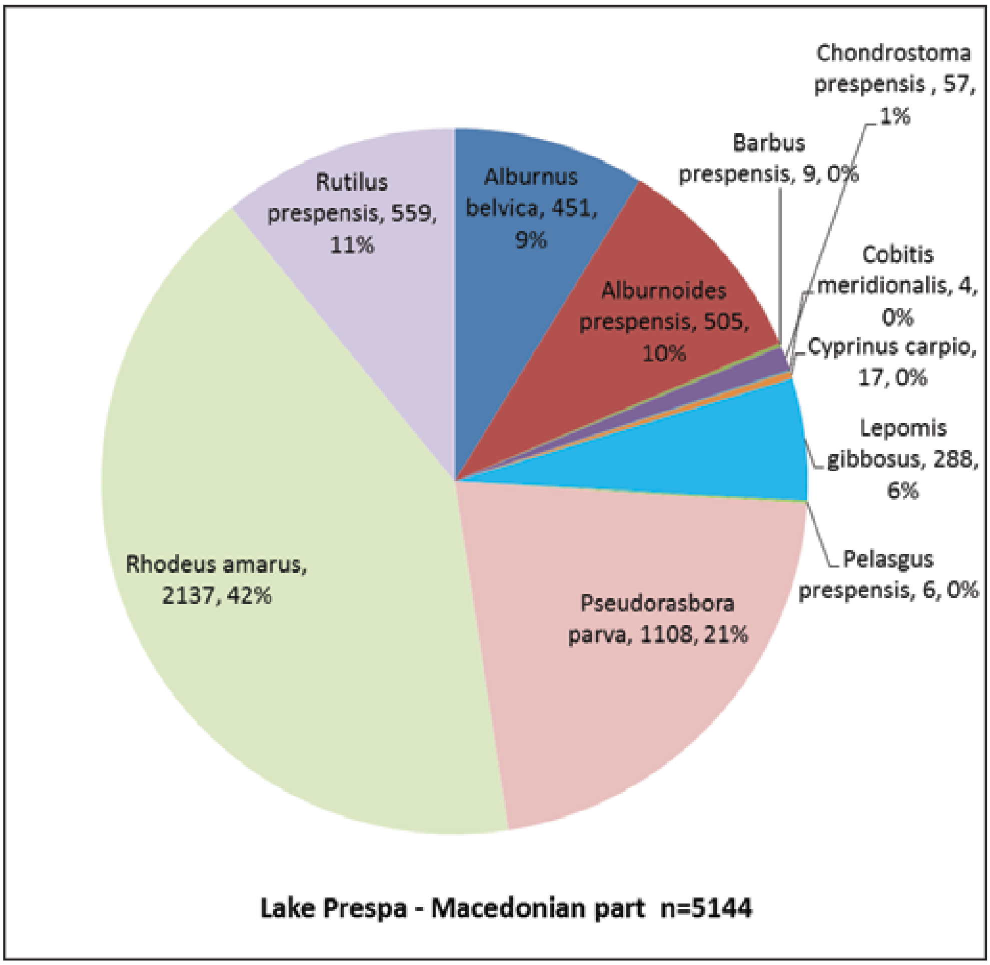


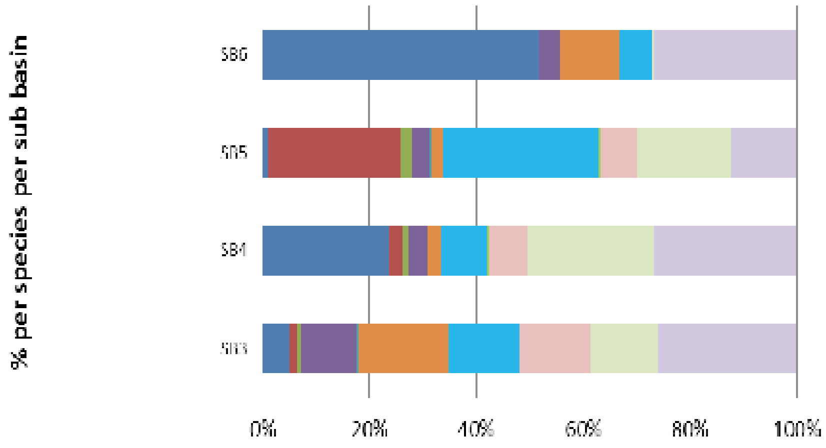
Figure 25. Relative and absolute fish species composition represented in the total catch at the Macedonian Part of Lake Prespa during the sampling campaign in October 2013

The total catch from all of the sub basins consisted of 12 caught species from 20 present at Lake Prespa. Their total number was 5144 specimens. As it can be seen from the Figure 25 above, the most dominant appears to be the three alien species bitterling, stone moroko and pumpkinseed respectively with total of 69% contribution.

The native species are represented with 8 of them with total abundance of only 31% all together, where most abundant are roach, spirling and bleak with more or less than 10% each of the total catch.

The rest of the native and endemic species are with low numbers from which nase (undermouth) is represented with 57 individuals, carp with 17 individuals and barbell with only 9. The rest of the native species caught were Prespa minnow with 6 and spine loach with only 4 individuals. It worth to mention, like a presence of a species for Prussian carp that 1 specimen was caught in the net but during the lifting of the net it escaped, and as not measured it isn't considered in any of the estimations. So, it can be stated that 12 species were registered during the sampling campaign in October 2012 at Lake Prespa at the Macedonian part. The commercially valued fish which are usually dominant like bleak, roach and carp, have very low presence in the MMG catch.

4.3.2.1.2 **CPUE**



	SB3	SB4	SB5	SB6
Alburnus belvica	1.76	32.60	3.18	146.84
Alburnoides prespensis	0.41	3.7	59.59	0.27
Barbus prespensis	0.24	1.53	5.52	
Chondrostoma prespensis	3.54	5.05	8.12	12.16
Cobitis meridionalis	0.08		0.34	
Cyprinus carpio	5.7	3.07	5.48	30.98
Lepomis gibbosus	4.48	12.18	71.35	17.52
Pelasgus prespensis		0.15	0.32	
Pseudorasbora parva	4.47	9.94	16.68	0.33
Rhodeus amarus	4.29	32.80	42.82	0.99
Rutilus prespensis	8.73	36.90	30.46	76.08

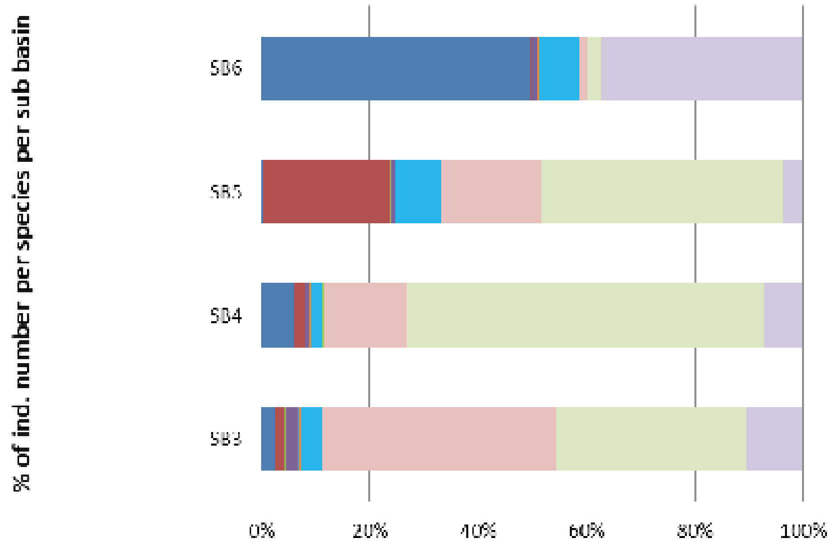
g/m²

Figure 26. CPUE expressed in biomass (g/m²) in percentage per species of total catch per sub-basin (BPUE) at Macedonian Part of Lake Prespa during the sampling campaign in October 2013

The CPUE in this case expressed through biomass per square meter of net, shows biggest values at SB5 Konjsko, where also biggest number of specimen 1957 was caught.

Furthermore in separate chapters are described each of the sub basins at the Macedonian part.

In the case of CPUE expressed in number of individuals (NPUE) per square meter of net, the bleak commercially valued fish, in the period when the sampling was performed, is mostly present in the open waters of the lake, whilst the alien species are mainly occupying the area close to the shore.



	SB3	SB4	SB5	SB6
Alburnus belvica	0.05	0.13	0.01	0.30
Alburnoides prespensis	0.03	0.05	0.84	0.00
Barbus prespensis	0.00	0.00	0.01	0.00
Chondrostoma prespensis	0.04	0.02	0.02	0.01
Cobitis meridionalis	0.01	0.00	0.00	0.00
Cyprinus carpio	0.01	0.01	0.01	0.00
Lepomis gibbosus	0.07	0.05	0.30	0.05
Pelasgus prespensis	0.00	0.00	0.01	0.00
Pseudorasbora parva	0.82	0.35	0.67	0.01
Rhodeus amarus	0.65	1.50	1.61	0.02
Rutilus prespensis	0.20	0.17	0.14	0.23

ind./m²

Figure 27. CPUE expressed in number of individuals/m² in percentage per species of total catch per sub-basin (NPUE) at Macedonian Part of Lake Prespa during the sampling campaign in October 2013

4.3.2.1.3 **SB3 - Asamati**

The Sub Basin 3 near the village of Asamati located at the Northeast part of Lake Prespa, regarding the fish fauna composition sampled in October 2013 shows presence of 10 species in the total catch of this campaign of 1271 individuals. From Fig. 25 dominance of the alien species can be clearly seen, whilst the presence of the native and endemic species to Lake Prespa is scarce. In fact, 82% of the fish number belongs to the three aliens: stone moroko, bitterling and pumpkinseed.

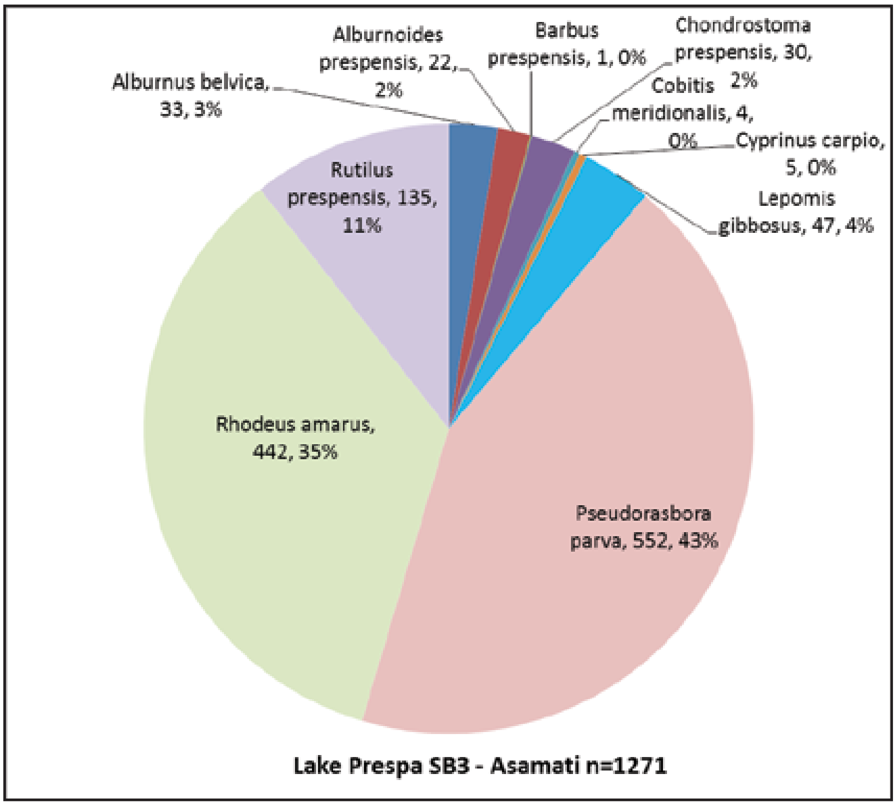
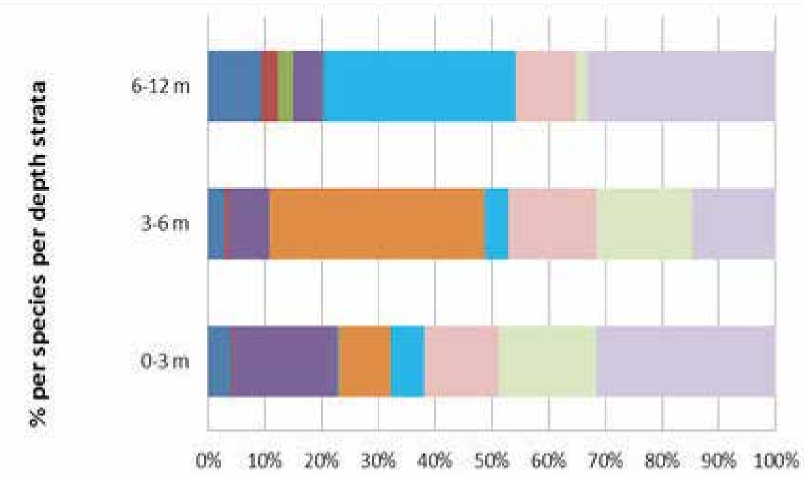


Figure 28. Relative fish species composition in total catch at SB3 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 5 individuals)

Having in mind the surface area of the lake that was covered within the sampling protocol in the SB3 in this period, the habitat constitution (reed belt=>submersed vegetation on sandy and muddy bottom) and also the seasonal distribution of the fish, shows quite distinction in presence of alien versus native species. It is also evident from Fig. 25 above, as in absolute as well in relative values. But, regarding the occupation of the depth strata stone moroko is abundant in all three within the range 0-12 m, which is followed by almost same relative abundance of bitterling.



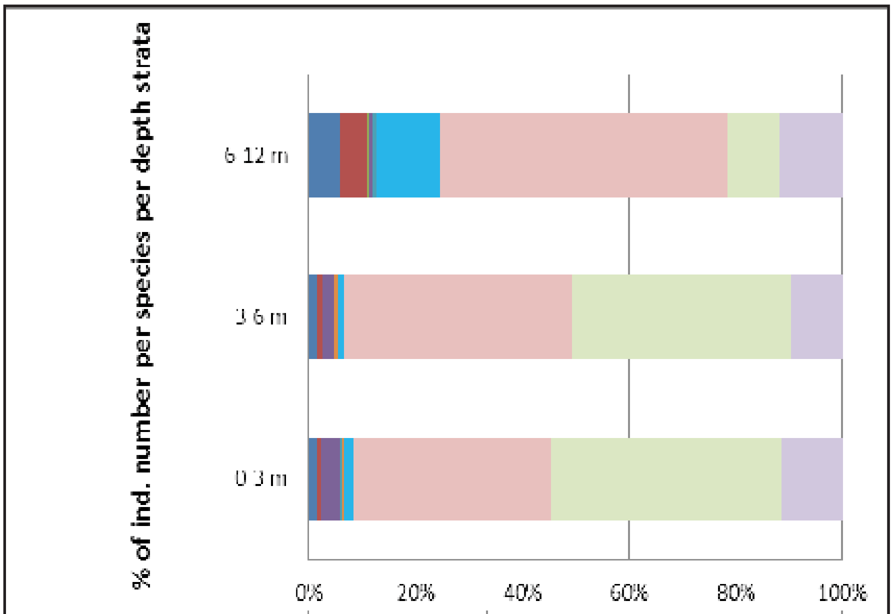
	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.47	0.36	0.93
Alburnoides prespensis	0.04	0.08	0.29
Barbus prespensis	0.00	0.00	0.24
Chondrostoma prespensis	2.16	0.87	0.52
Cobitis meridionalis	0.04	0.00	0.04
Cyprinus carpio	1.06	4.63	0.00
Lepomis gibbosus	0.70	0.49	3.28
Pseudorasbora parva	1.53	1.90	1.04
Rhodeus amarus	2.04	2.05	0.21
Rutilus prespensis	3.70	1.79	3.24

gr/m²

Figure 29. CPUE expressed in biomass (g/m²) in percentage per species of total catch per depth strata (BPUE) at SB 3 locality

Regarding the CPUE interpreted in relative biomass expressed in g/m² of nets catchable area (675 m²) which usually should serve for fish stock and fishing quota estimations, as shown in Fig. 27 above, the dominance of the native roach and carp is higher due to their natural characteristics - larger bodies compared to those of stone moroko and bitterling.

In the case of CPUE expressed in number of individuals (NPUE) per square meter of net from the following figure clearly can be seen the dominance of the alien species, mostly stone moroko and bitterling.

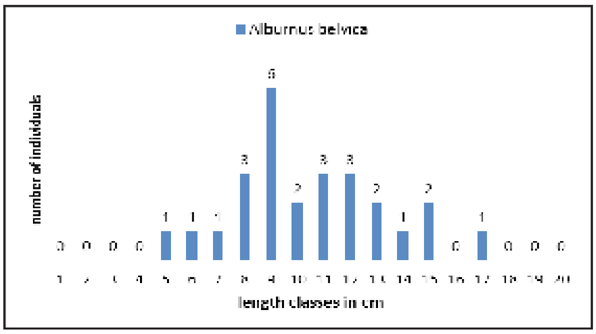


	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.03	0.04	0.07
Alburnoides prespensis	0.01	0.02	0.07
Barbus prespensis	0.00	0.00	0.00
Chondrostoma prespensis	0.07	0.06	0.01
Cobitis meridionalis	0.01	0.00	0.01
Cyprinus carpio	0.01	0.01	0.00
Lepomis gibbosus	0.03	0.08	0.15
Pseudorasbora parva	0.72	1.06	0.67
Rhodeus amarus	0.83	1.01	0.12
Rutilus prespensis	0.22	0.24	0.15

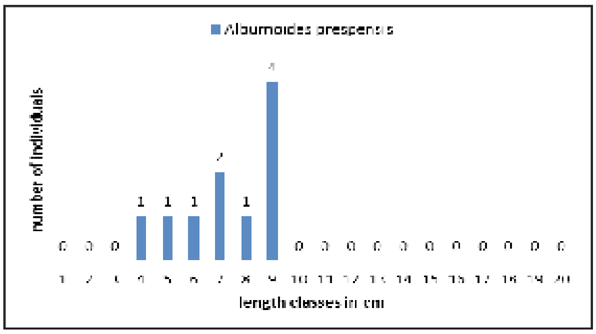
ind./m²

Figure 30. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 3 locality

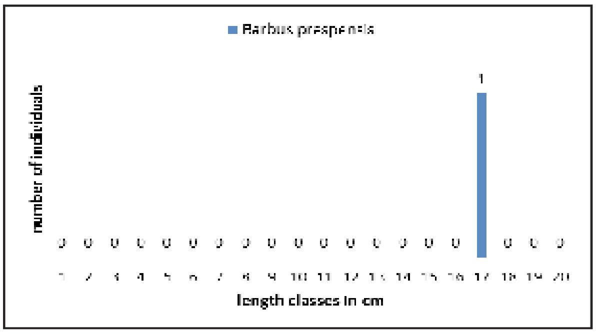
The length frequency distributions of the fish species caught during the survey at the SB 3 are presented in the following figure (a-i).



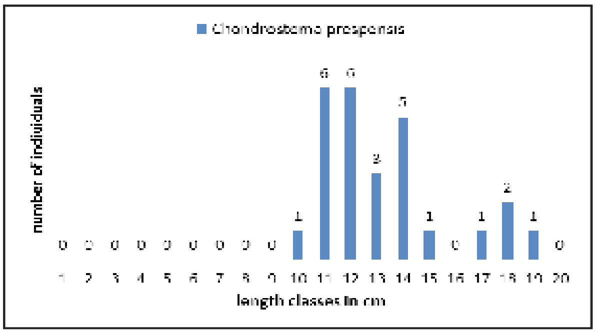
a)



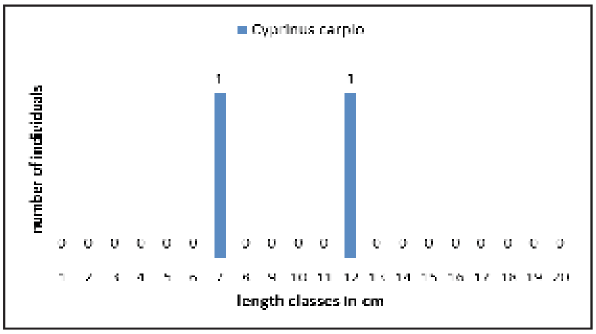
b)



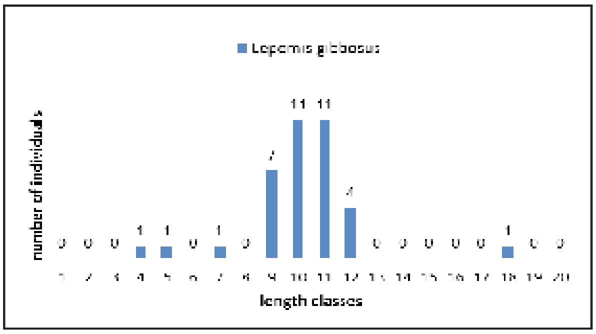
c)



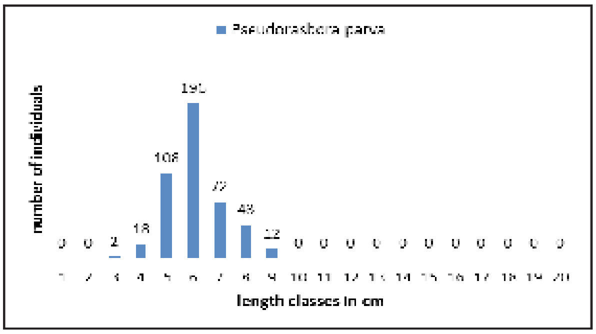
d)



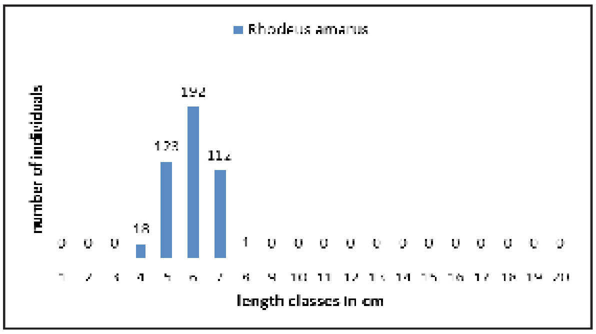
e)



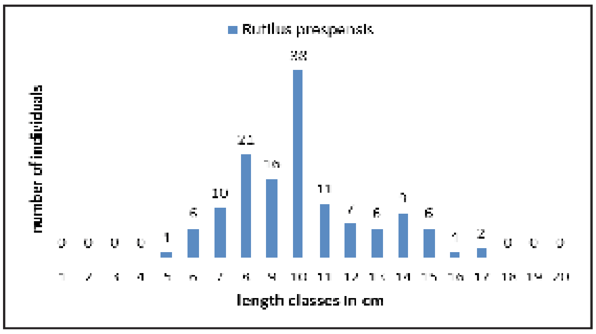
f)



g)



h)



i)

Figure 31. Length classes frequency per species for the SB 3 Asamati (from a to i)

At this sub basin 3, large frequency in length clases can be observed among the native species like roach with 13 classes which are in a range 3 year cohorts from 0+ to 2+, than the bleak with 12 classes with almost same age classes like the roach, and the nase with 9 classes with yearly cohorts of 1+ and 2+.

Among the alien specis, the pumpkinseed is represented with 8 length classes with several youngs of the year and several adults. The stone moroko and the bitterling according their natural comon size, are represented with small individuals with 6 and 4 classes respectively.

4.3.2.1.4 **SB4 – Otesevo**

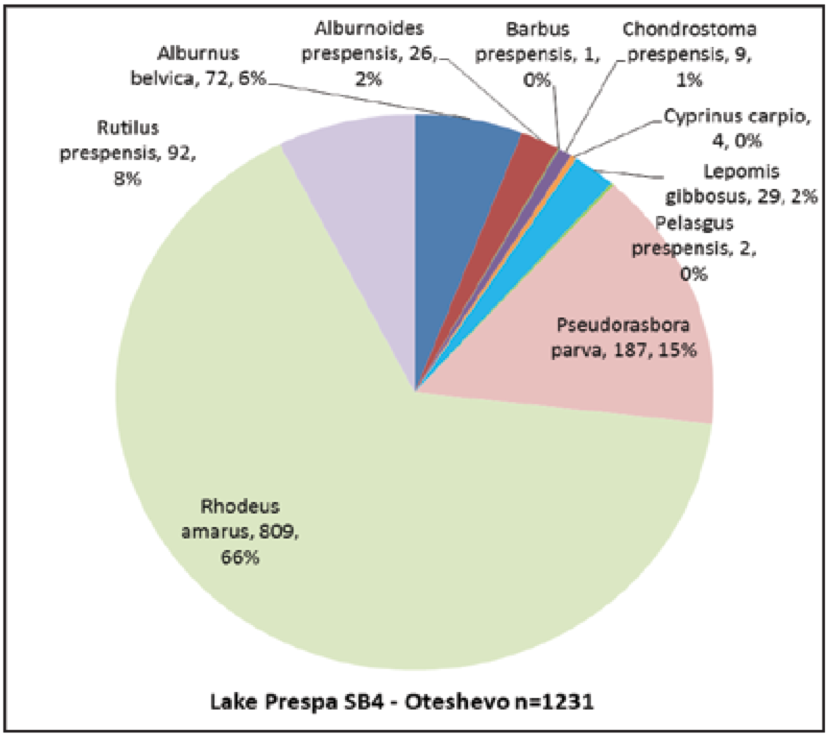
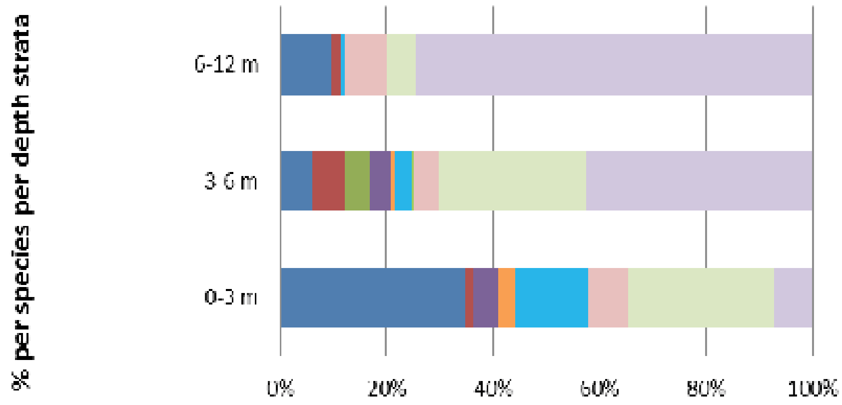


Figure 32. Relative fish species composition in total catch at SB4 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 5 individuals)

The Sub Basin 4 near the vicinity Otesevo located at the Northwest part of Lake Prespa, regarding the fish fauna composition sampled in October 2013 shows presence of 10 species in the total catch of this campaign of 1231 individuals. From the above Fig. 32 dominance of the alien species can be clearly seen, whilst the presence of the native and endemic species to Lake Prespa is drastically lower. In fact, 83% of the fish number are the three aliens: bitterling, stone moroko and pumpkinseed respectively. Thus, in this sub basin the bitterling is dominant with 66% of the total specimens. In this case the 7 native species are present with only 17%, where 8% are occupied by roach and 6 with bleak.

Having in mind the surface area of the lake that was covered within the sampling protocol in the SB4 in this period, the habitat constitution (reed belt=>partly rocky=> submersed vegetation on sandy and muddy bottom) and also the seasonal distribution of the fish, shows quite distinction in presence of alien versus native species.



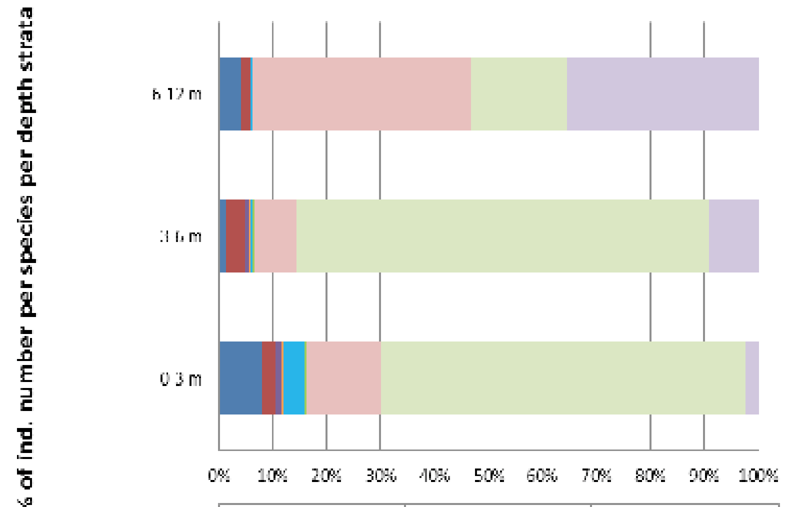
	0-3 m	3-6 m	6-12 m
Alburnus belvica	28.43	2.09	2.08
Alburnoides prespensis	1.13	2.19	0.37
Barbus prespensis	0.00	1.53	0.00
Chondrostoma prespensis	3.64	1.42	0.00
Cyprinus carpio	2.79	0.28	0.00
Lepomis gibbosus	10.84	1.14	0.20
Pelasgus prespensis	0.04	0.10	0.00
Pseudorasbora parva	6.15	1.76	1.73
Rhodeus amarus	22.05	9.63	1.12
Rutilus prespensis	5.86	14.87	16.16

g/m^2

Figure 33. CPUE expressed in biomass (g/m^2) in percentage per species of total catch per depth strata (BPUE) at SB 4 locality

Regarding the CPUE interpreted in relative biomass expressed in g/m^2 of nets which usually should serve for fish stock and fishing quota estimations, as shown in Fig. 33 above, the dominance of the native roach and bleak is higher due to their natural characteristics - larger bodies compared to those of stone moroko and bitterling.

Regarding the CPUE expressed in number of individuals (NPUE) per square meter of net but, regarding the occupation of the depth strata within the whole range 0-12 m the bitterling highly dominated in this case. The second dominant is almost equally distributed but in much lower number. Interesting case is the presence of the bleak in the shallow zone 0-3 m.

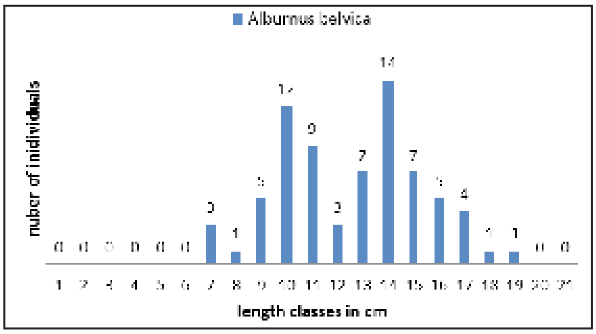


	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.10	0.01	0.01
Alburnoides prespensis	0.03	0.02	0.00
Barbus prespensis	0.00	0.00	0.00
Chondrostoma prespensis	0.01	0.00	0.00
Cyprinus carpio	0.01	0.00	0.00
Lepomis gibbosus	0.05	0.01	0.00
Pelasgus prespensis	0.00	0.00	0.00
Pseudorasbora parva	0.17	0.05	0.09
Rhodeus amarus	0.82	0.52	0.04
Rutilus prespensis	0.03	0.06	0.08

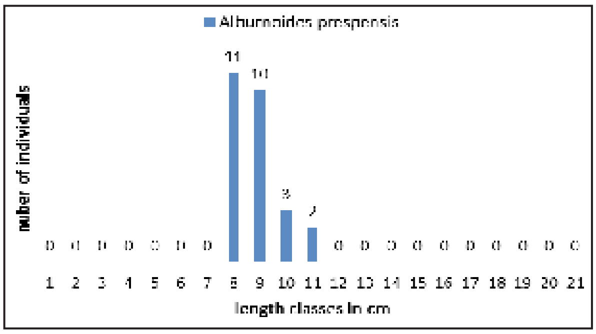
ind./m^2

Figure 34. CPUE expressed in number of individuals/m^2 in percentage per species of total catch per depth strata (NPUE) at SB 4 locality

The length frequency distributions of the fish species caught during the survey at the SB 4 are presented in the following figure (a-j).



a)



b)

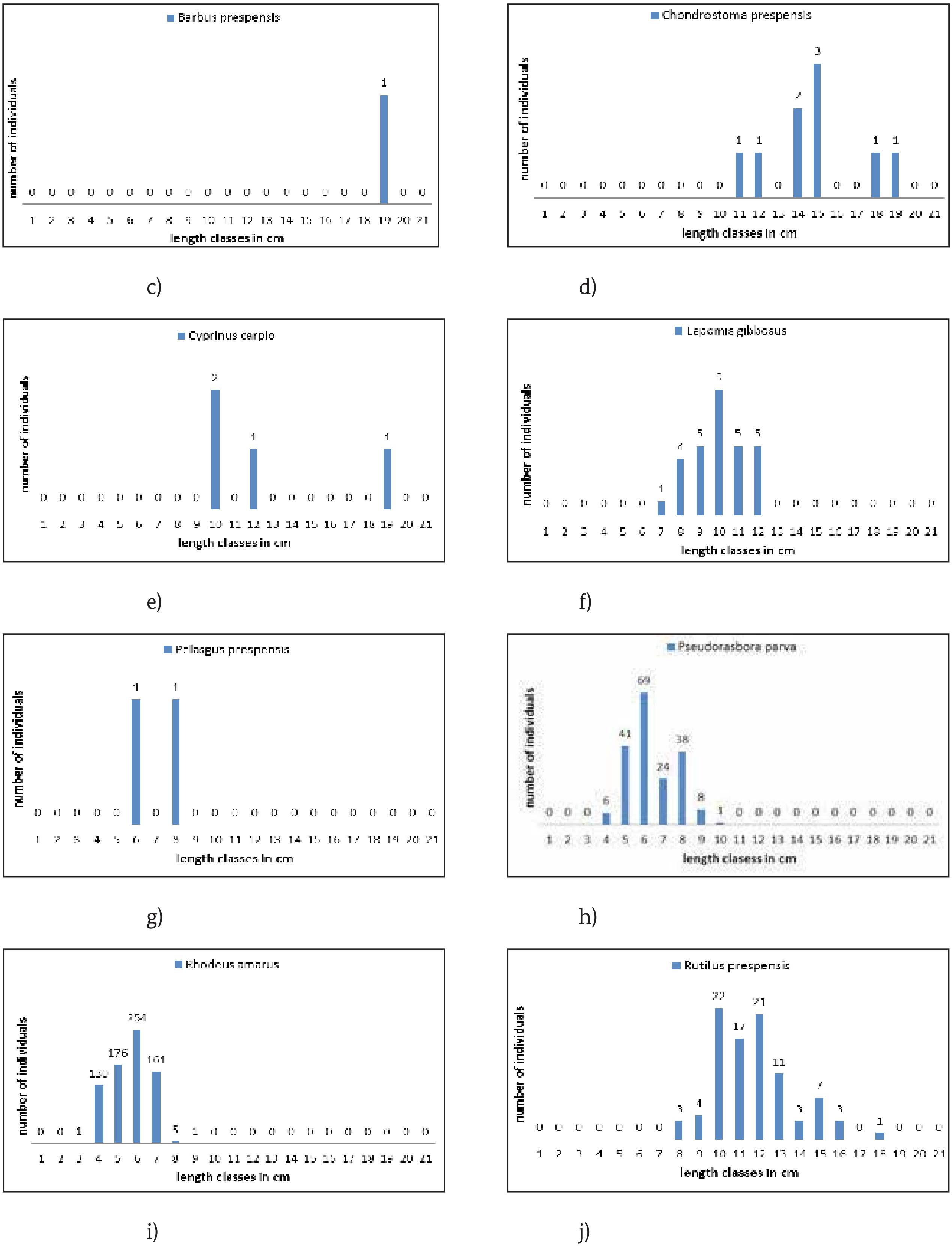


Figure 35. Length classes frequency per species for the SB 4 Otesevo (from a to j)

At this sub basin 4, large frequency in length classes can be observed among the native species like roach with 13 classes which are in a range 3 year cohorts from 0+ to 2+, than the bleak with 12 classes with almost same age classes like the roach, and the nase with 9 classes with yearly cohorts of 1+ and 2+.

Among the alien specis, the pumpkinseed is represented with 8 length classes with several youngs of the year and several adults. The stone moroko and the bitterling according their natural comon size, are represented with small individuals with 6 and 4 classes respectively.

4.3.2.1.5. **SB5 – Konjsko**

The Sub Basin 5 near the village Konjsko located at the Western part of Lake Prespa, regarding the fish fauna composition sampled in October 2013 shows presence of 11 species in the total catch of this campaign of 1957 individuals.

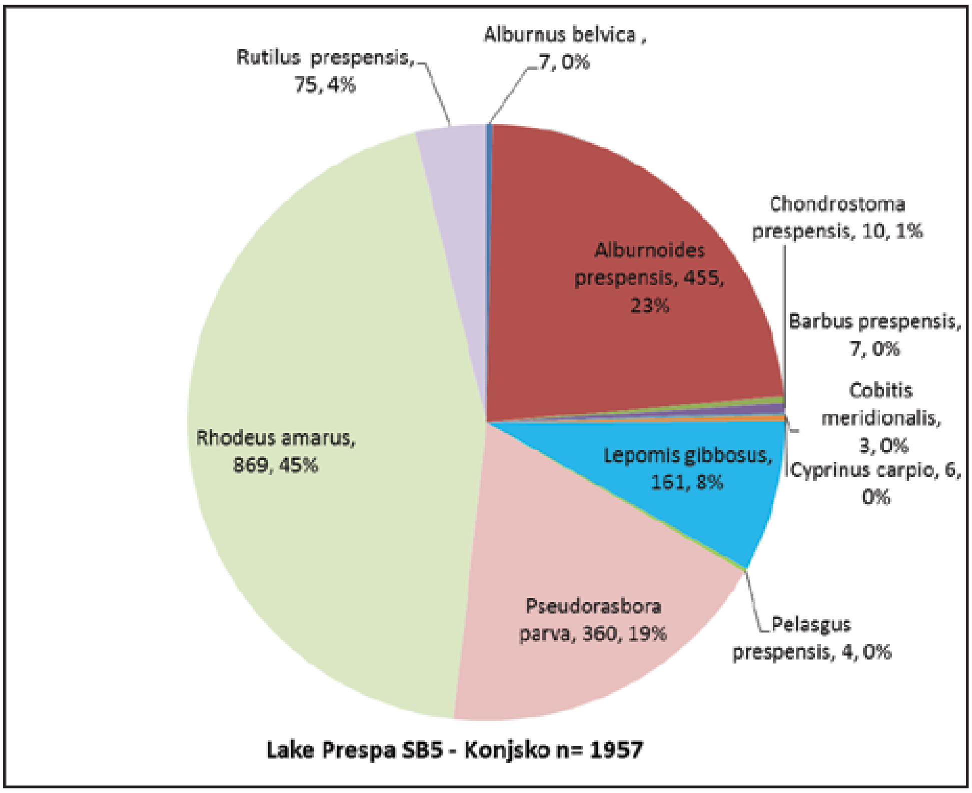
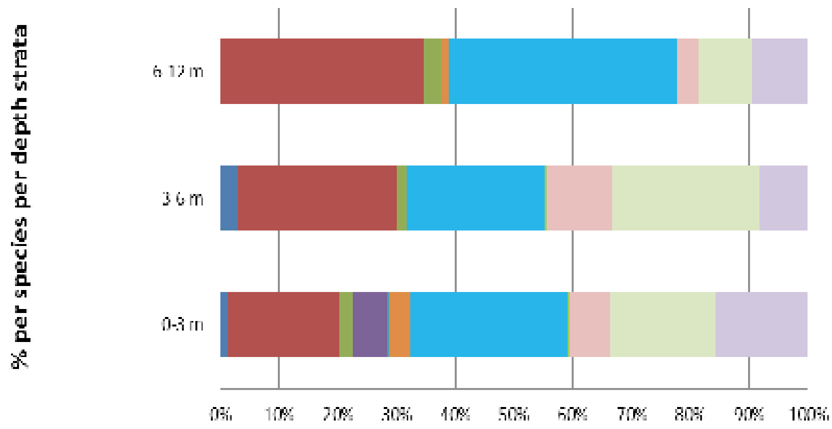


Figure 36. Relative fish species composition in total catch at SB5 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 7 individuals)

From the above Fig. 36 dominance of the alien species can be clearly seen in this case also but with a bit lower participation of 72% with respectful dominance of bitterling, stone moroko and pumpkin seed in this case.

The presence of the native and endemic species to Lake Prespa of 28% is consisted of prespa spirling with 23% and roach with 4%, whilst the rest natives are represented negligibly with few specimens, which from other hand shows the diversity richness of this sub basin if included the 1 specimen of Prussian carp that escaped from the net during lifting the net.



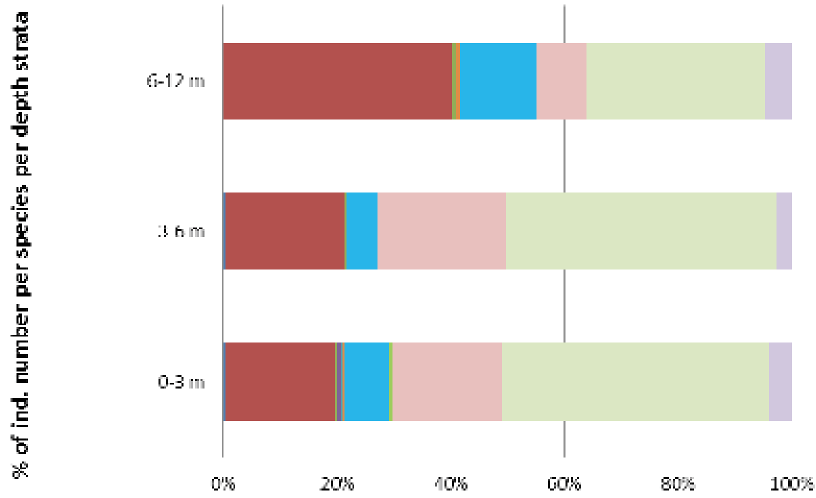
	0-3 m	3-6 m	6-12 m
Albumus belvica	1.77	1.42	0.00
Albumoides prespensis	25.29	13.89	20.42
Barbus prespensis	3.13	0.78	1.61
Chondrostoma prespensis	8.12	0.00	0.00
Cobitis meridionalis	0.34	0.00	0.00
Cyprinus carpio	4.64	0.00	0.84
Lepomis gibbosus	36.39	12.08	22.88
Pelasgus prespensis	0.25	0.07	0.00
Pseudorasbora parva	8.98	5.71	1.99
Rhodeus amarus	24.49	12.90	5.44
Rutilus prespensis	20.82	4.08	5.56

g/m^2

Figure 37. CPUE expressed in biomass (g/m^2) in percentage per species of total catch per depth strata (BPUE) at SB 5 locality

The CPUE interpreted in relative biomass expressed in g/m^2 of nets, as shown in Fig. 37 above, in the biomass in this sub basin 5 are dominating again the alien species also in biomass with pumpkinseed and bitterling. From the natives the biomass is mainly consisted of spirlin (shneider) and roach.

Bearing in mind the habitat constitution (reed belt on a rocky substrate=>rocky and gravel=> submersed vegetation on sandy and muddy bottom) and also the seasonal distribution of the fish, shows quite distinction in presence of alien versus native species. From Fig. 38 below, regarding the occupation of the depth strata within the whole range 0-12m most of the species are evenly distributed.

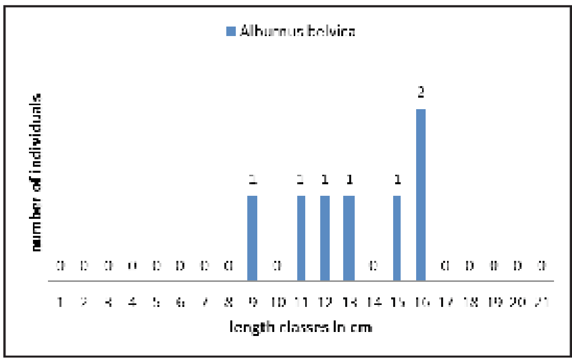


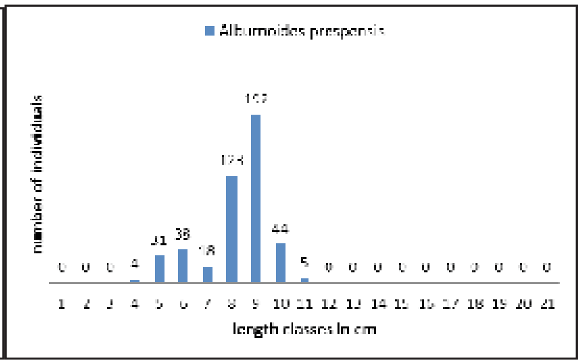
	0-3 m	3-6 m	6-12 m
Alburnus belvica	0.02	0.01	0.00
Alburnoides prespensis	0.95	0.79	0.75
Barbus prespensis	0.01	0.01	0.02
Chondrostoma prespensis	0.04	0.00	0.00
Cobitis meridionalis	0.01	0.00	0.00
Cyprinus carpio	0.02	0.00	0.01
Lepomis gibbosus	0.39	0.21	0.25
Pelasgus prespensis	0.02	0.00	0.00
Pseudorasbora parva	0.96	0.85	0.17
Rhodeus amarus	2.31	1.81	0.58
Rutilus prespensis	0.20	0.10	0.09

ind./m²

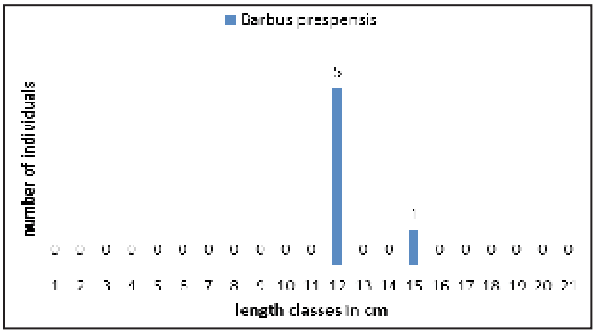
Figure 38. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 5 locality

The length frequency distributions of the fish species caught during the survey at the SB 5 are presented in the following figure (a-j).

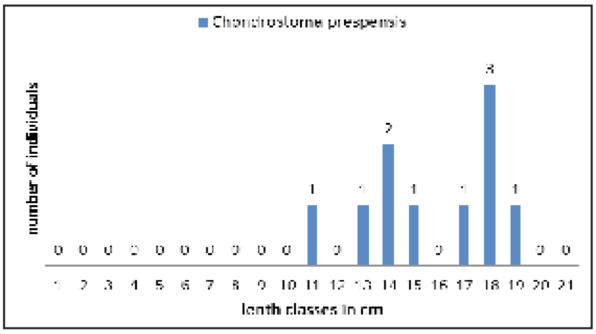




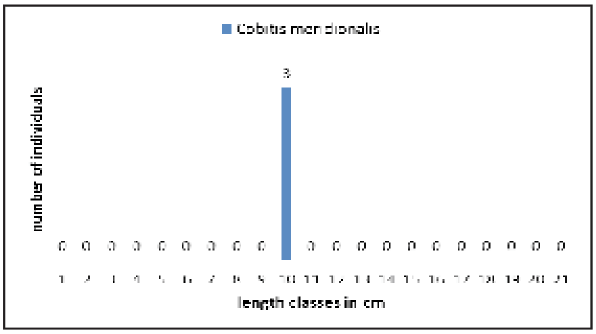
a) b)


Barbus prespensis
number of individuals
length classes in cm

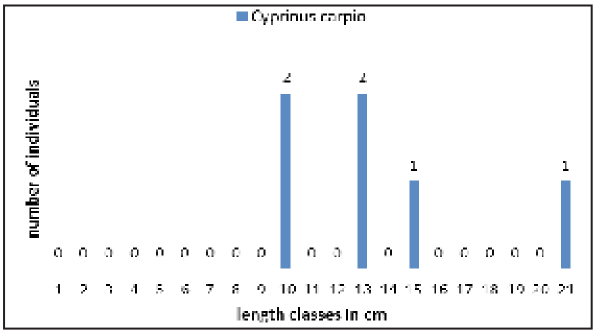
c)


Chondrostoma prespensis
number of individuals
lenth classes in cm

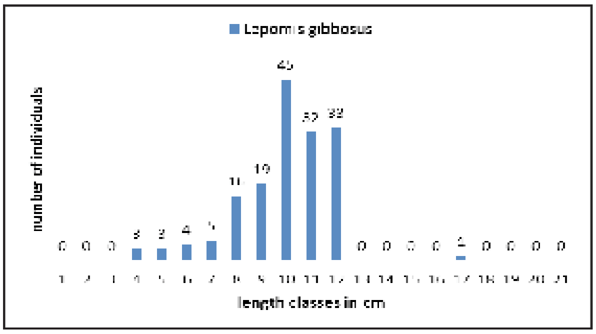
d)


Cobitis meridionalis
number of individuals
length classes in cm

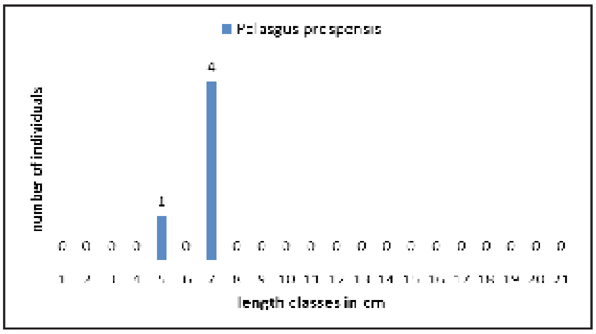
e)


Cyprinus carpio
number of individuals
length classes in cm

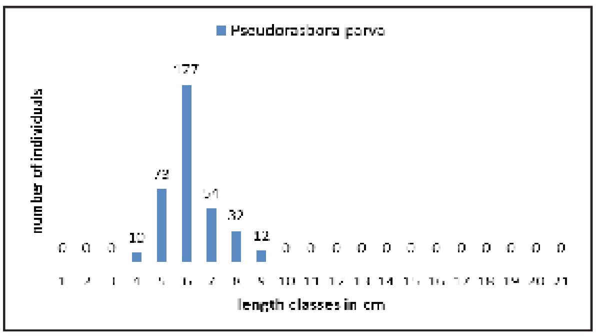
f)


Lepomis gibbosus
number of individuals
length classes in cm

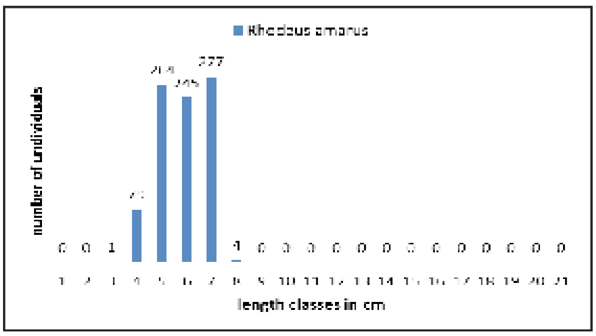
g)


Pelasgus prespensis
number of individuals
length classes in cm

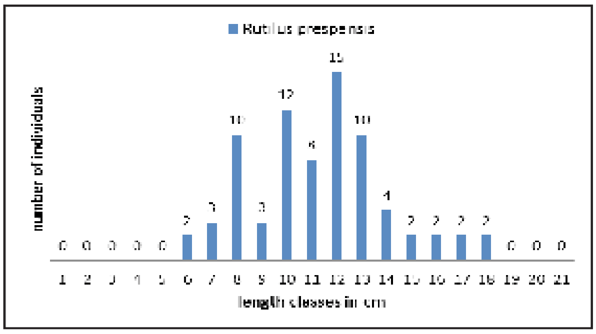
h)


Pseudorasbora parva
number of individuals
length classes in cm

i)


Rhodeus amarus
number of undividuals
length classes in cm

j)



k)

Figure 39. Length classes frequency per species for the SB 5 Konjsko (from a to k)

At this sub basin 5, large frequency in length clases can be observed anly among the native roach with 13 classes which are in a range 3 year cohorts from 0+ to 2+. Second comes the prespa spirling with 8 classes in a range of 0+ and 1+ years, than prespa nase with 7 length classes although almost with one specimen per class but ranging from 1+ and 2+ years.

Among the alien species, the pumpkinseed is represented with 9 length classes with several youngs of the year and significant number of adults. The stone moroko and the bitterling according their natural comon size, are represented with small individuals with 6 and 4 classes respectively but in a huge number compared to the rest, especially the second one mentioned.

4.3.2.1.6. **SB6 – Central Plate**

The Sub Basin 6 located at the central pelagic part of Lake Prespa, regarding the fish fauna composition sampled in October 2013 shows presence of 8 species in the total catch of this campaign of 685 individuals.

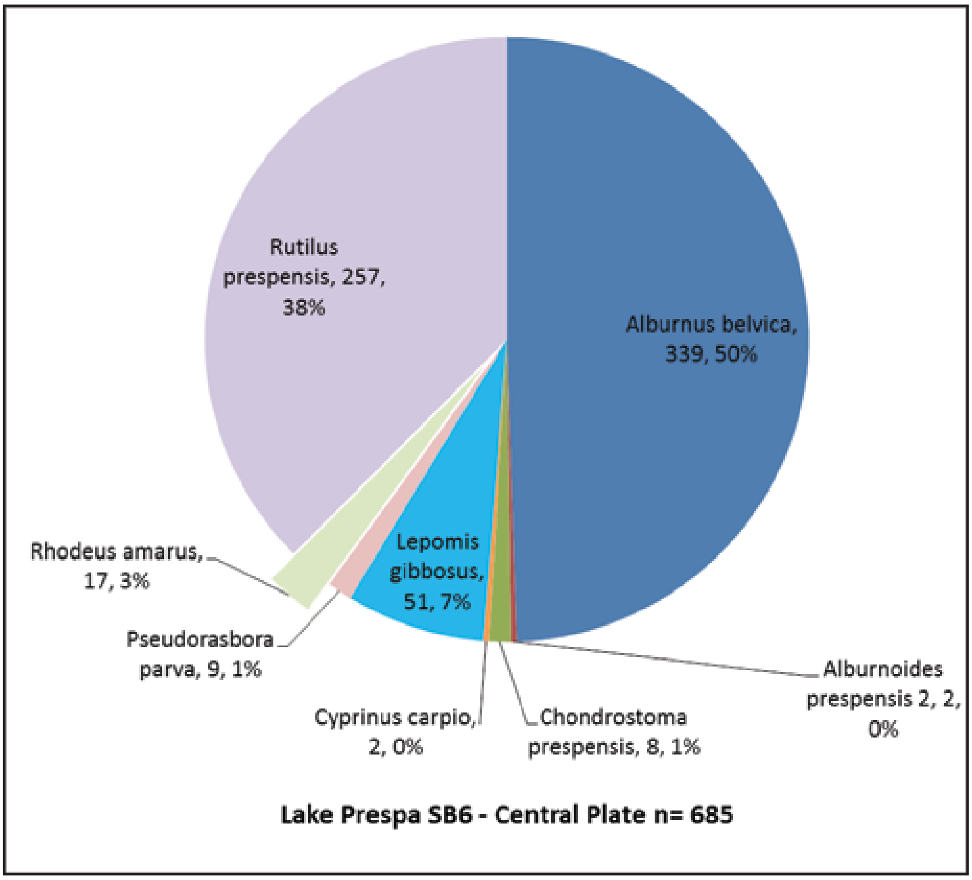


Figure 40. Relative fish species composition in total catch at SB6 in the sampling campaign October 2013 (Note: Data label of 0% are representing species whose total number was less than 2 individuals)

From the above Fig. 40 unlike the rest of the sub basins the dominance has been switched into favor of the native species versus the aliens. In fact the natives were present with 89% from which the bleak with 50% and the roach with 38 and the rest of 1% comprised the nase, spirlin and carp, respectively.

The 11% of the aliens were belonging to pumpkinseed (7%) bitterling (3%) and stone moroko (1%).

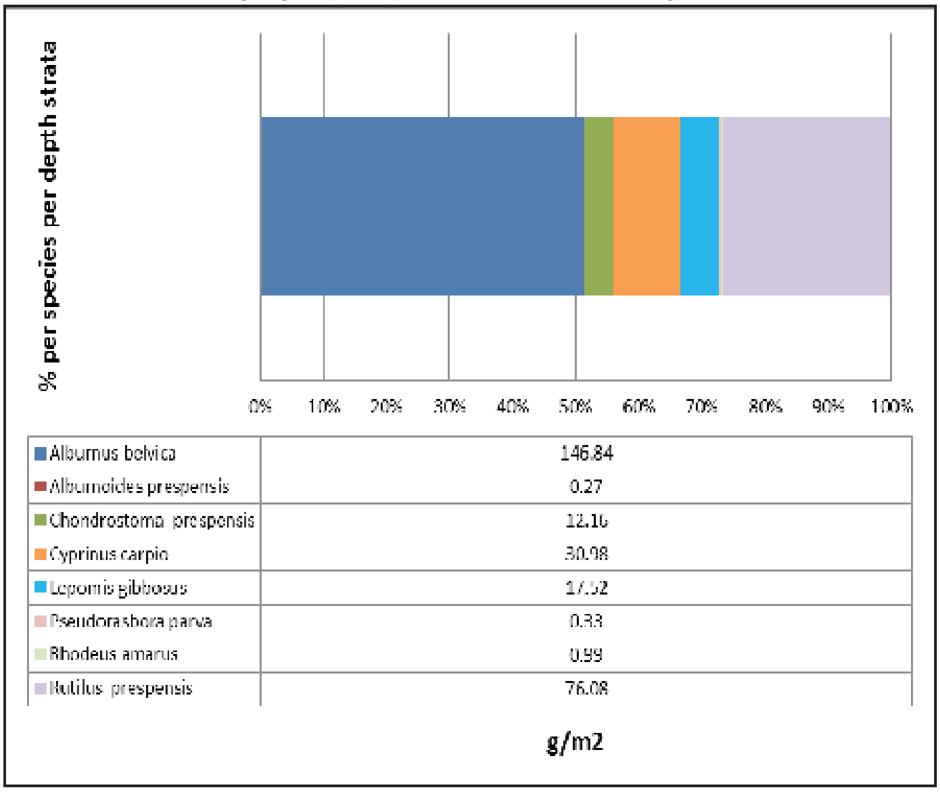


Figure 41. CPUE expressed in biomass (g/m^2) in percentage per species of total catch per depth strata (BPUE) at SB6 locality

In the case of the Central Plate sub basin, unlike the rest of those close to the shore, the CPUE in biomass corresponds to the species composition as in relative as well as in absolute values.

Regarding the CPUE expressed in number of individuals (NPUE) per square meter of net species pattern follows that from BPUE.

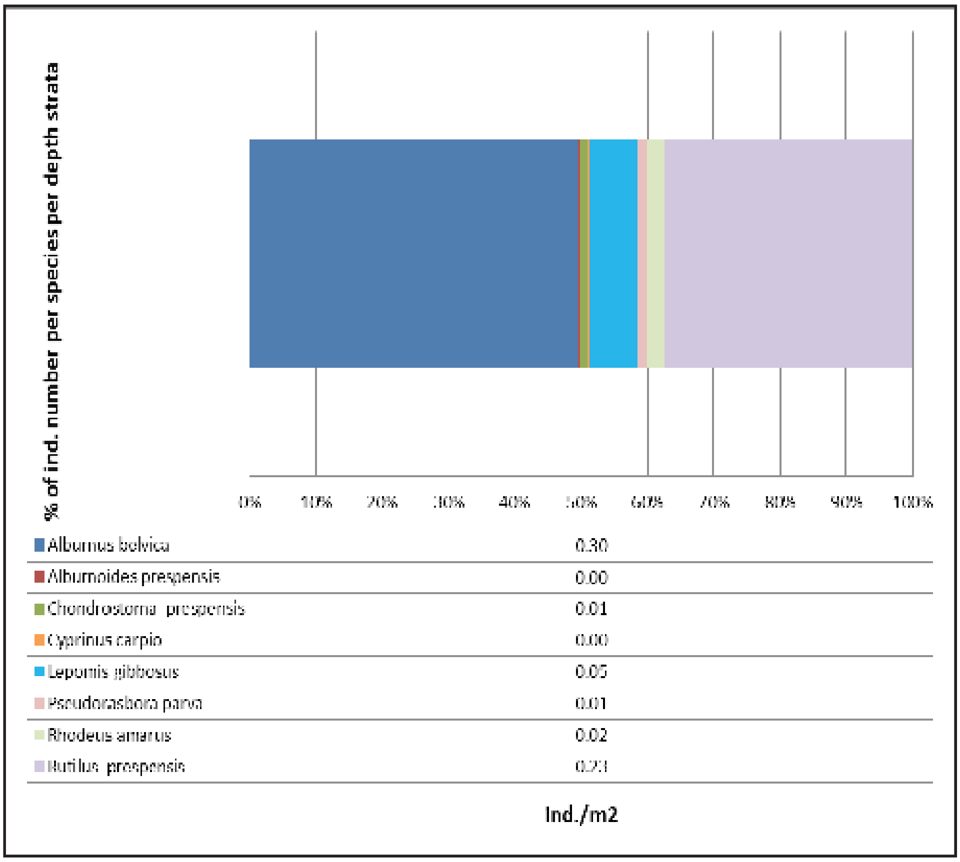
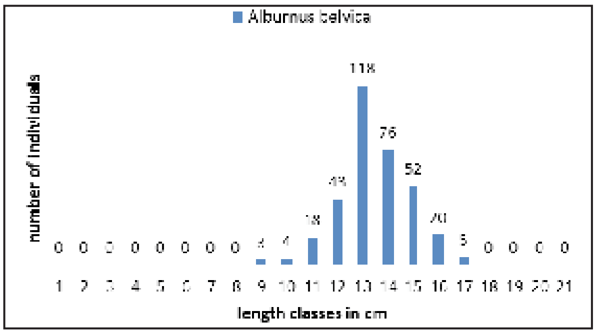
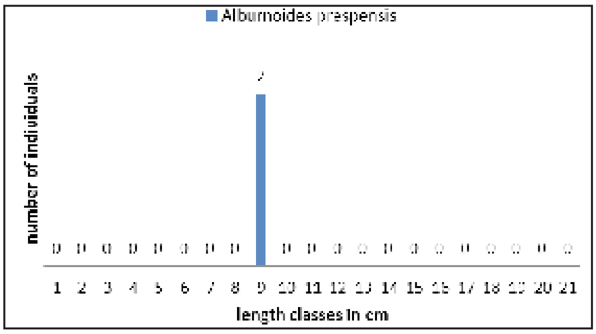


Figure 42. CPUE expressed in number of individuals/m² in percentage per species of total catch per depth strata (NPUE) at SB 6 locality

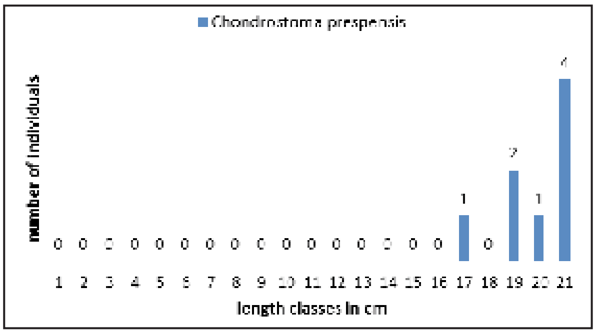
The length frequency distributions of the fish species caught during the survey at the SB 6 are presented in the following figure (a-h).



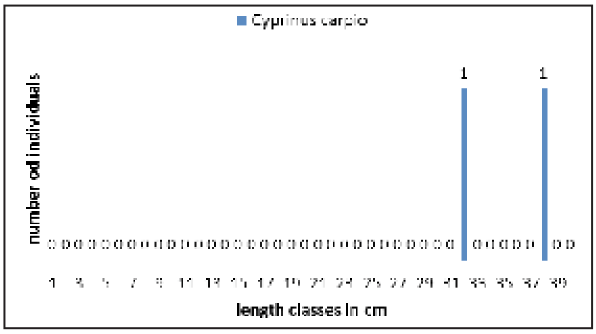
a)



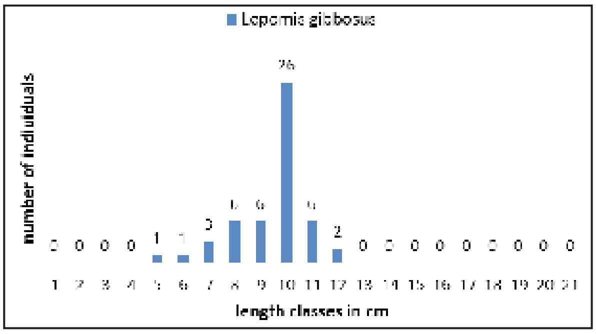
b)



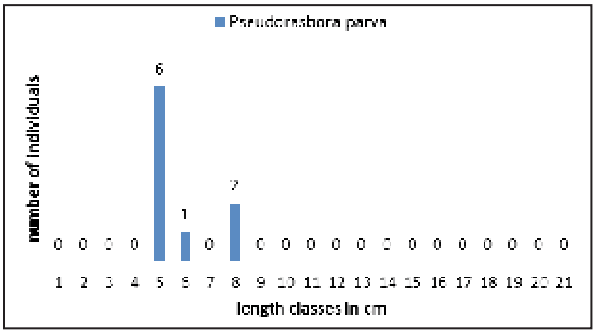
c)



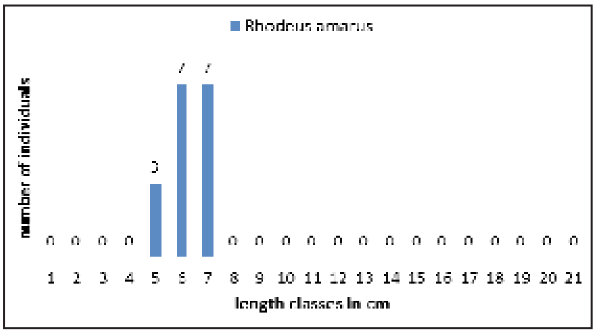
d)



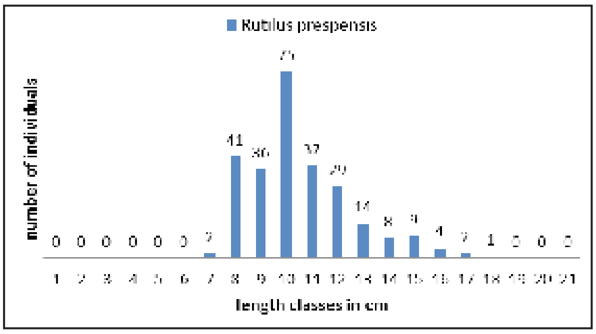
e)



f)



g)



h)

Figure 43. Length classes frequency per species for the SB 6 Central plate (from a to h)

Central plate has depth range of 12-16 meters and is representing pelagic region of Lake Prespa which is also 2/3 of the lake's bottom. Species composition of this sub-basin from this sampling campaign is closest to the fishery statistics for this part of the year.

4.3.3. **Synthesis for Lake Prespa**

Depending of the fish species distribution per sub-basins according to similarity and dissimilarity of the habitat conditions, it varies from 1.04 (SB4) to 1.67 (SB2). The data shows higher species abundance in the littoral sampling sub-basins 0-12 m – SB1-SB5 with exception of SB4 due to large and very dense composition of the reed belt along the shore and in depths bigger than 3m. The values of this index presented in the below figure are proportional with the number of fish species and their abundance number present in the given sub-basins, thus as higher is the number of species, the values are higher.

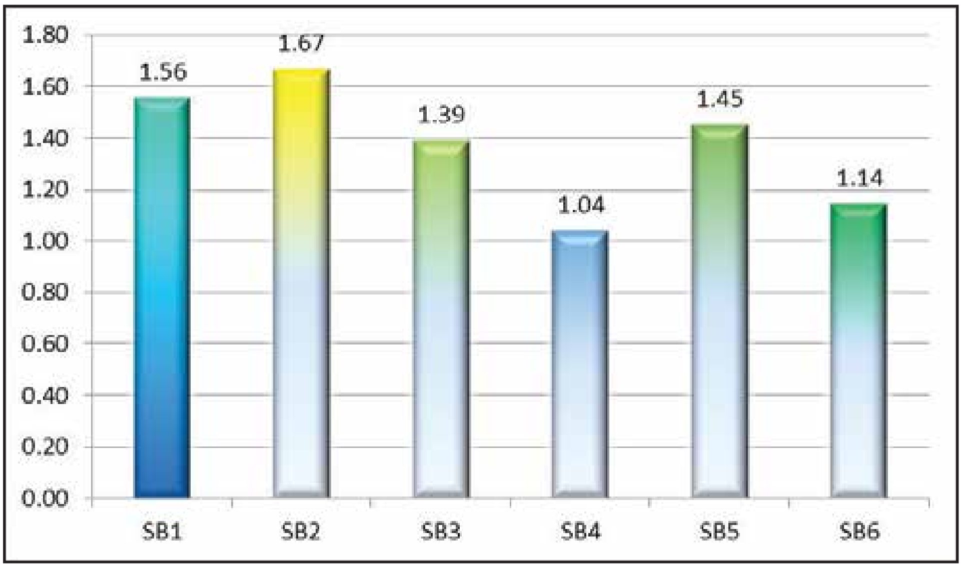


Figure 44. Biodiversity (Shanon-Wiener) index of the six sampled sub-basins of the water body Lake Prespa

For the species evenness variation index is the opposite case, data presented in the below figure are inverse with the number of fish species and their abundance number present in the given sub-basins, thus as higher is the number of species, the values are lower.

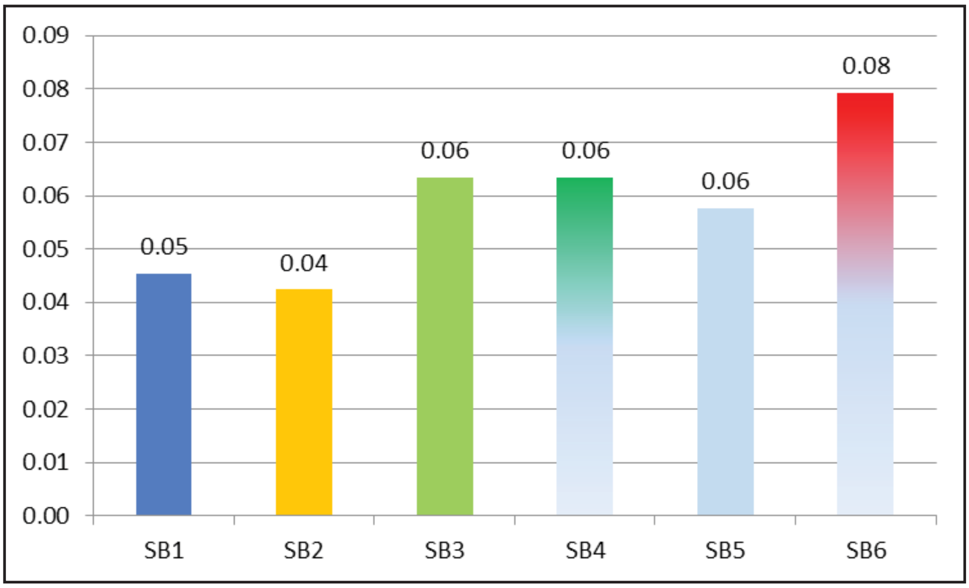


Figure 45. Evenness variation index represented fish species in the six sampled sub-basins of the water body Lake Prespa

4.4. **Discussion of results trans-boundary sampling scheme**

Following the distribution of fish species in the various depth strata of the lake, during survey in entire lake area and strata, multi-mesh gillnets were randomly set in a range of depths as required by the EN 14757 standard. This provided information on the distribution of fish in each sub basin of the lake for the sampling campaign in October 2013.

It is clear from the analysis of the data for each fish species in relation to depth that the greatest concentrations of alien species occurred in the shallower depths, while the most economic valuable species like bleak, roach and carp at the pelagic part of the lake. Hence, this kind of sampling as in type as well in the period performed is just showing small part of the fish distribution – assemblage and condition.

From this sampling campaign 15 fish species were recorded at Lake Prespa from which at SB1 and SB2 15, 12 at SB5, 10 at SB3 and SB4 and 8 at SB6, comparing to Tab. 1, 75% of inhabiting fish fauna.

Bleak is endemic to Prespa Lake. It is the most commercial one and shows stability in Lake Prespa. This species is major prey of fish eating birds and is also a target species of fishermen and local people. Thanks to its life-history strategy it can cope with such high predation mortality. On both sides of the lake it is the dominant economic resource in terms of fishery.

Based on the data from this sampling campaign presence of bleak (from 4% at the depth strata 0-3 m and 13% at the depth strata 6-12 m) comparing with previous records (Crivelli et alt., 2006) reveals better species status in entire stock on Albanian side of the Lake.

The most valuable commercial fish in Lake Prespa is carp. It's the most important commercial species at the transboundary area. But, during this sampling campaign at the Macedonian part only 17 specimens were caught from all-in total 5144 individuals of all species.

Other sensitive species Prespa barbell and Prespa nase are also less distributed with hypotheses that it is indicative of a failure in the reproduction or development due to water quality and water oscillation with direct impact on species spawning grounds as riverine and gravel habitats.

The Prespa sprilin shows a tendency of satisfied presence (from 5-11%, particularly at the depth strata 0-3 m and 3-6 m). In case of an accelerated eutrophication obviously this species will face unpredictable problems due to the fact that primary this species is generally a riverine species with adaptations to this standing water body where it spawns also.

Prespa roach was widely spread through all sampling sub-basins and represented with highest number of length classes (13) among all native and alien species.

Bitterling and stone moroko are introduced species and there are with no commercial value. Both species have a negative impact on native species (e.g. Prespa spirlin), that need to be tested. This species is much more numerous in Micro Prespa than in Macro Prespa, especially from the beginning of this century.

Following the increased presence of alien species in terms of number of individuals of bitterling, stone moroko, pumpkinseed etc, the role as aliens in relation to natives in Prespa Lakes is poorly understood. The WFD status class has been discussed with a number of people and it seems to be general agreement that the presence of one or more established alien species is certainly not 'natural' and is likely to affect the WFD status class in a detrimental way.

The pumpkinseed in the data is well present and based on experiences and personal communication with local fisherman it has increased in Prespa Lake. During this and previous MMGN surveys it was evident from the local fishermen that caught specimens of this species were thrown back in to the lake and according to their endurance they survive almost all.

Other recorded species like tench seems to not establish stable populations in Prespa Lake. During this sampling campaign this species was caught only at Albanian side.

The unique set of the fish populations inhabiting this basin together with unique specifications of the Prespa Lakes Basin urges with no doubt to more severe measures in maintaining the freshwater biodiversity.

Being an intensive agricultural area Prespa Lakes watershed presence of additional nutrient load as well as other agrochemicals, combined with lake's habitats disruption and high fishing pressure are representing the main threats to the fish biodiversity. In addition presence of the high number of alien fish species and fish eating predatory birds leads towards intervention as in the local strategies for biodiversity protection as well in the legislative part in the countries shearing the lake.

5 Conclusions

EN 14757 standard with MMGN was for the first time performed on Lake Prespa and results from the analysis of the data for each fish species in relation to depth are showing that the greatest concentrations of alien species occurred in the shallower depths, while the most economic valuable species like bleak, roach and carp at the pelagic part of the lake.

Stock development cannot be registered and discussed or drawing any conclusions due to lack of commercial fishing on the Macedonian side of the Lake (within the last seven years) which represents the biggest part of Lake Prespa.

Fish as indicators of water quality under WFD - In the areas with higher nutrient load at the Macedonian Part of the lake mainly alien species were present with high abundance. Bitterling and stone moroko were the most abundant followed by stone moroko.

Species biodiversity and how this is affected by pressures - At present Prespa Lake fish fauna is presented with 20 fish species (Table 1. 13 native and 7 alien) from which 15 were sampled during the CSBL sampling campaign October 2013, from which 10 were native and 5 alien species. From the present ones in the Lake Prespa fish fauna, remained uncaught are 3 native species: eel, stone loach and one of the minnows – *Phoxinus lumaireul* and two alien species: catfish and mosquito fish. The most abundant native species in this sampling campaign were bleak, roach and spirlin and the most abundant alien species were bitterling, stone moroko and pumpkinseed.

The main extraction of the fish as biomass is by the fish eating birds (cormorants and pelicans). More than 60 tons per year mainly bleak (more than 90%) are extracted by cormorants. (UNDP, 2012) The biggest nesting colony in Europe is situated on the island Golem Grad - Macedonia with more than 2000 breading pares.

Till 2007, fishery represents the biggest pressure on fish fauna at Lake Prespa. Since 2007 there is no legal fishing on Macedonian side (2/3 of Lake Prespa belong to Macedonia). According to different personal communications this kind of ilegal fishing is quite significant in the fish biomass extraction.

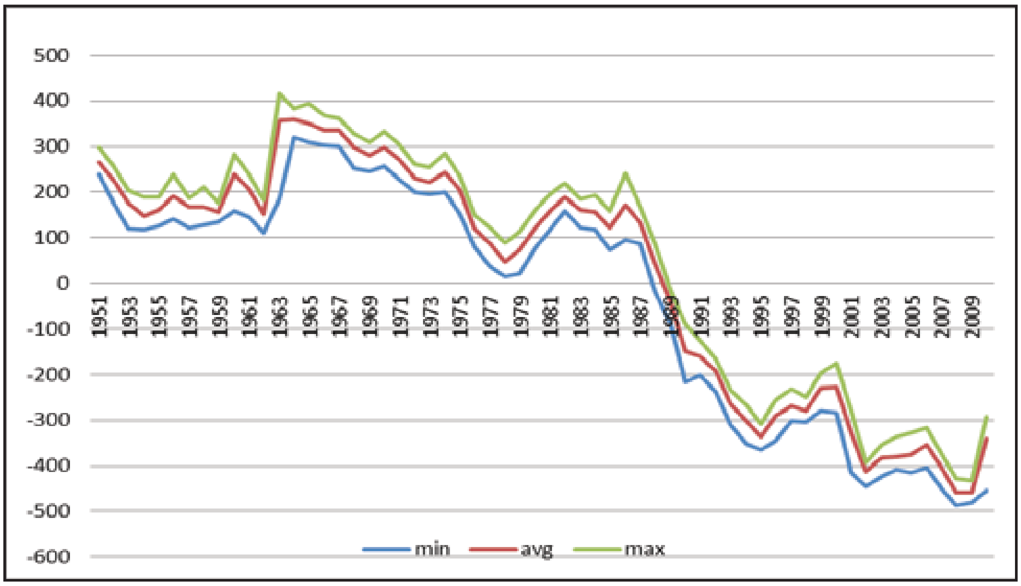
The following table of Total allowable fish catch quotas was produced during the “Transboundary Fish and Fishery Management Plan for Prespa Lakes Basin” Project in 2011 in which the leading institution was PSI Hydrobiological Institute - Ohrid.

Annual TAFCQ for commercial fishery (kg)			
Species	Macedonian part	Albanian part	Greek part
Cyprinus carpio (carp)	35.000	25.000	25.000
Squalius prespensis (chub)	6.000	3.000	3.000
Chondrostoma prespense (nase)		1.000	1.000
Rutilus prespensis (roach)	15.000	7.000	8.000
Alburnus belvica (bleak)	170.000	200.000	60.000
TOTAL	226.000	236.000	97.000
All other alien species	Unlimited	Unlimited	Unlimited

Annual TAFCQ for recreational fishery (kg)			
Species	Macedonian part	Albanian part	Greek part
Cyprinus carpio (carp)	4.000	1.000	2.000
Squalius prespensis (chub)	3.000	500	1.000
Chondrostoma prespense (nase)		200	200
Rutilus prespensis (roach)	6.000	3.000	1.500
Alburnus belvica (bleak)	18.000	5.000	5.000
TOTAL	31.000	12.700	9.700
All other alien species	Unlimited	Unlimited	Unlimited

The same Project gives data about the relatively large density of fish ecto and endoparasites among the native species and especially bleak and roach, which from other hand are dominant in biomass. Such kind of density gives relative knowledge of abundance of these two species in the Lake which correlates their infestation. The recorded presence of new fish parasite species reflects the influence of cormorant colony to the fish fauna in another sense.

- Main eutrophication source in the Prespa Lakes watershed is agriculture with high nutrient load and other harmful substances.
- High number of alien fish species is another pressure to the native fishes due to the spawning grounds inter-competition and overlap of the ecological niche.
- Fluctuations of the Prespa Lake water level with shrinkage of the lake surface is affecting spawning habitats and changes in the water quality.



Sustainable use of fish stocks and fisheries – Trilateral implementation of Fishery Management Plan for Prespa Lake should be performed followed by legislation harmonization among transboundary countries.

The present monitoring survey and data collected serve as a prove for the usefulness of applying standardized lake fish parameters to assess the ecological status of lakes and support the development of integrated monitoring of entire lake. Further to that following European experiences development and setting ecological quality based on different metrics.

Applied protocol and sampling scheme used within CSBL MMGN sampling in future should be widened as in fishing gears and other fish surveillance devices as well for time series.

6 Recommendations

Lake Prespa is a shared resource, and no action can be taken by one country without impacting the resources and conditions in the other country.

It is of utmost importance to re-establish and to reactivate the bilateral co-management authority ("Lake Prespa Fisheries Authority or Commission"), which already existed in the previous century to manage the fisheries and related resources. Any other experiences similar to the conditions of Lake Prespa fish fauna and its ecosystem are welcomed and appreciated. Representatives from national institutions, local authorities, fishermen's organizations, research institutions, civil society etc. are recommended to be considered for membership.

These mix authority (technical and political) could be establish in the frame of *The Agreement between Albanian and Macedonian Governments for the Protection and Sustainable Development of Lake Prespa and its Watershed*, signed by respective prime ministers some years ago. According to this agreement the riparian countries will take the necessary measures, amongst others, to protect the biodiversity (particularly endemic species), to ensure the sustainable use of natural resources, and to prevent and control the economic activities from seriously damaging (and polluting) the environment. In the light of the above possible measures it is very important that the fishing effort be reduced to conserve biodiversity and to restore the balance of the underwater fauna in order to exploit the available resources in a sustainable manner.

Lake Prespa fish fauna concerned as a world heritage deserves adequate research resources for its protection and human wellbeing.

- Joint Monitoring of fish stock and spawning grounds and habitats is one of necessary important actions.

- Prespa Lake Fish stock assessment based on time series using all necessary fishing gears and other surveying technics.

- Proposed measures and actions from previous above-mentioned Project for Lake Prespa fish with respect to improvement of fisheries management, protection of biodiversity of fish fauna and lowering pressure on fishes remains same (UNDP, 2012):

No	Measures	Actions
1.	Trilateral Fishery Management	Establishing Joint Prespa Fishery Commission (JPFC)
2.	Monitoring of the water quality, fish stock monitoring	Establishing local monitoring stations in the three countries in cooperation with scientific institutions and other relevant stakeholders
3.	Joint Monitoring Technical Protocol	Quality Assurance and Data Acquisition (Designated responsible implementing bodies)
4.	Improved Fish Statistics	Implementing unique software (Data exchange) Establishing Fishery Data Base
5.	Fish Stock Assessment	Integrated actions (open cross border expeditions and surveillances with joint resources) FSA Revision and relevant changes of the actual Fishing Master Plans for Prespa Lake and Prespa Lake Watershed on the Macedonian side.
6.	Physical guarding of the fish stocks	Establishing national guarding bodies (state and private)
7.	Conservation	Conservation action plans per fish species Total ban on Prespa barbell for 6 years period Total ban on Prespa trout for 3 years period Stocking program only with autochthonous fish related to specific habitats
7.	Alien fishes combat	Selective and ameliorative fishing
8.	Fishing limits	Determining the allowable smallest catchable size per species Determining the spawning periods and close fishing season per species
9.	Spawning grounds - habitats	Defining strict natural fish spawning grounds (where any activities without special permission of the national management bodies and JPFC are allowed) Improving the conditions of spawning grounds (entrances in to the rivers from the lake for Prespa nase)
10.	Catch quotas	Determining of Annual Total Allowable Fish Catch Quotas (ATAFCQ) per country / per lake / per species
11.	Fishing regulations	Maximum allowed fishing gears and fishing equipment for commercial and recreational fishery
12.	Fish Stocking	Designing of Joint Fish Stocking Program (JFSP) based on obtained fish spawning individuals from the lakes or streams

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8 Attachments

ANNEX I. Presence of certain economically important fish species in the annual fish catch in the Prespa Lake (Source: Riboprespa – former concessioner)

Year	Fish species (%)						
	Carp (%)	Prespa nase (%)	Bleak (%)	Roach (%)	Others (barbell/ chub) 1990-2007		Total annual fish catch (kg)
1946	30.02	15.50	18.86	26.83	7.76		115,272
1947	27.21	29.58	28.03	10.37	4.79		99,229
1948	27.21	34.42	18.31	10.56	2.13		135,888
1949	15.89	21.19	11.81	27.04	24.04		137 ,385
1950	41.05	24.97	12.55	16.10	5.33		143,052
1951	38.56	36.68	17.67	6.38	0		138,308
1953	28.78	40.69	14.41	0	16.13		130,926
1954	31.33	38.18	14.86	10.27	5.36		131,656
1955	52.03	8.28	20.35	0	19.34		138,138
1958	25.27	24.60	34.30	15.83	0		115, 205
1959	13.30	53.44	18.98	14.29	0		93,716
1960	15.58	46.66	21.42	16.34	0		127,423
1961	10.79	46.37	20.20	22.63	0		151,053
1962	10.99	48.16	31.79	9.05	0		148,2 06
1963	19.71	34.76	33.33	12.20	0		173, 416
1964	11.46	32.94	35.91	19.69	0		173,405
1965	12.11	31.10	33.21	23.58	0		165,281
1966	20.49	32.84	34.45	12.22	0		149,837
1967	18.08	37.64	29.09	15.19	0		143,566
1968	11.20	41.75	27.99	19.05	0		126,427
1969	16.97	46.09	20.31	16.62	0		108,136
1970	13.57	43.92	15.12	27.38	0		121,070
1971	3.76	41.77	29.18	23.27	2.02		80,643
1972	8.88	54.48	13.84	20.78	2.02		102,918
1973	9.99	53.80	6.34	27.28	2.59		97,911
1974	2.92	65.13	17.11	11.79	3.05		82,733
1975	0.59	68.50	26.93	1.20	2.78		87,635
1976	0.09	72.80	21.52	2.76	2.84		88,802
1977	0.39	52.26	14.70	5.12	27.53		118,344
1978	0.23	55.64	19.01	2.08	22.98		126,530
1979	0.29	41.09	30.75	9384	18.02		113,339
1980	0.11	47.87	27.52	12.49	12.02		86,005
1981	0.04	35.47	36.99	3.61	23.90		57,926
1982	0.14	47.35	40.45	2.53	9.53		84,148
1983	0.05	37.95	39.36	0	22.64		53,394
1984	0.13	33.17	39.19	5.15	22.36		77,958
1985	0.05	12.31	49.74	3.28	34.63		69,902
1986	0.06	10.01	60.62	20.25	9.06		52,747
1990	0.15	7.69	78.08	5.19	0.47	8.42	69,389
1991	0.05	2.47	77.14	8.84	0.32	11.17	75,994
1992	0.15	2.57	81.64	9.34	0.47	5.82	82028
1993	0.60	2.89	82.69	5.97	0.74	7.11	83,814
1994	2.18	4.77	65.18	7.17	0.53	2.01	83,252
1995	0.56	2.11	80.42	7.02	0.17	9.72	64,687
1996	1.18	3.80	75.54	3.67	0.30	15.52	57,272
1997	1.54	5.68	72.41	4.21	0.67	16.12	28,516
1998	0.22	4.49	80.24	1.51	0.72	12.82	30,365
1999	56.77	10.50	27.36	0.35	0.41	4.61	7,131
2000	54.08	19.49	6.10	0	0	20.33	11,547

2001	100	0	0	0	0	0	3,040
2002	0.30	16.69	41.12	0	0	41.88	0,659
2004	9.30	6.59	57.11	0	0	27.01	107,317
2005	2.45	2.64	69.42	0	0	25.48	47,001
2006	1.16	1.83	90.74	0	0	6.27	17,997
2007							18,582

ANNEX II. Preliminary fish sampling scheme

Albania (Macro and Micro Prespa)												
Method	Jan.	Feb.	Mar.	Apr.	May	Jun.	July	Aug.	Sep.	Oct.	Nov.	Dec.
Benthic multi-mesh gillnets European Standard							Macro Prespa Kallamas Micro Prespa Al-Gr border					
Pelagic multi-mesh gillnets European standard												
Fyke Net							Macro Prespa Kallamas Micro Prespa Al-Gr border					
Electro-fishing transects												
Larval Trap							Macro Prespa					
Beach Seine							Macro Prespa Kallamas Micro Prespa Al-Gr border					
Catch data												

Macedonia (Macro Prespa)												
Method	Jan.	Feb.	Mar.	Apr.	May	Jun.	July	August	Sep.	Oct.	Nov.	Dec.
Benthic multi-mesh gillnets European Standard							Asamati Oteshevo Konjsko					
Pelagic multi-mesh gillnets European standard							Kazan					
Fyke Net								Asamati Oteshevo				
Electro-fishing transects								Asamati				
Larval Trap							Asamati Oteshevo Konjsko pelagial					
Beach Seine								Asamati Oteshevo Konjsko				
Catch data												

If there is a concessionaire for fishing

For SB1 Kallamas, SB3 Asamati and SB4 Otesevo beside the multimesh gillnets, **fyke nets** with wings of 5 and 10 mm mesh size from knot to knot were planned to be used.

Fish larval traps were planned to be used at SB1 Kallamas, SB2 Liqenas, SB3 Asamati and SB5 Konjsko as littoral sub basins as well as in SB6 Central plate as pelagic sub basin to cover presence of the species recruitment – 0^+ of cyprinid and salmonid fishes in Lake Prespa.

Two different mesh sizes (5 and 10 mm from knot to knot) of **beach seine nets** as more harmless fishing gear for the sampled individuals and more productive, were foreseen to cover the circadian peaks of fish species. This fishing gear was planned to be used at SB1 Kallamas, SB3 Asamati, SB4 Otesevo and SB5 Konjsko.

ANNEX III. Lake Prespa points of sampling and additional sampling data

LAKE PRESPA SUB BASIN 1 - KALLAMAS POINTS OF SAMPLING (POS) AT ALBANIAN PART

Net number	Sector's Sub Basin 1	Depth strata	Coordinates		Date	Time of net setting	Time of net lifting	Time efforts (min)	Sechi D (m)	T (oC)
1	F	0-3 m	40514346	20574044	17.10.2013	1728	630	782	2,7	16,4
2	F	0-3 m	40513890	20565284	17.10.2013	1738	640	782	2,7	16,4
3	A	0-3 m	40524736	20564460	16.10.2013	1720	610	810	2,8	16,6
4	E	0-3 m	40525410	20554977	18.10.2013	1725	630	785	3	16,2
5	D	0-3 m	40535410	20563551	16.10.2013	1735	630	815	2,8	16,6
6	F	0-3 m	40513520	20574144	19.10.2013	1725	620	815	2,8	16,5
7	G	0-3 m	40512623	20581618	19.10.2013	1735	630	815	2,8	16,5
8	C	0-3 m	40531662	20554389	18.10.2013	1740	645	810	3	16,2
9	F	0-3 m	40520060	20563482	20.10.2013	1740	620	800	2,8	16,4
10	E	0-3 m	40525259	20555077	20.10.2013	1730	435	665	2,8	16,4
11	K	0-3 m	40525648	20581430	17.10.2013	1810	710	780	2,7	16,4
12	E'	3-6 m	40522928	20570556	16.10.2013	1755	645	810	2,8	16,6
13	K'	3-6 m	40511501	20582212	21.10.2013	1800	640	760	2,5	16,1
14	B'	3-6 m	40502577	20580325	20.10.2013	1800	705	785	2,8	16,4
15	A'	3-6 m	40503992	20575929	20.10.2013	1810	715	785	2,8	16,4
16	G'	3-6 m	40524742	20581928	17.10.2013	1800	655	775	2,7	16,4
17	I'	3-6 m	40505708	20581614	19.10.2013	1800	700	780	2,8	16,5
18	C'	3-6 m	40512922	20581321	18.10.2013	1810	640	750	3	16,2
19	E'	3-6 m	40514646	20575435	18.10.2013	1820	655	755	3	16,2
20	J'	3-6 m	40504962	20581811	19.10.2013	1810	715	785	2,8	16,5
21	G'	3-6 m	40523158	20580720	16.10.2013	1810	700	810	2,8	16,6
22	H'	6-12 m	40531162	20583229	17.10.2013	1815	725	790	2,7	16,4
23	E'	6-12 m	40520071	20571957	16.10.2013	1820	715	815	2,8	16,6
24	K'	6-12 m	40521425	20575137	20.10.2013	1835	740	785	2,8	16,4
25	B'	6-12 m	40512698	20581302	20.10.2013	1855	755	780	2,8	16,4
26	A'	6-12 m	40511950	20582014	20.10.2013	1920	810	810	2,8	16,4
27	G'	6-12 m	40524969	20582127	17.10.2013	1825	740	795	2,7	16,4
28	I'	6-12 m	40511576	20582807	20.10.2013	1920	830	790	2,8	16,4
29	C'	6-12 m	40514496	20575634	18.10.2013	1855	730	795	3	14,2
30	E'	6-12 m	40515396	20575733	18.10.2013	1905	735	750	3	14,2
31	J'	6-12 m	40504962	20582109	21.10.2013	1825	705	800	2,5	16,1
32	G'	6-12 m	40522705	20575634	16.10.2013	1835	730	815	2,8	16,6

LAKE PRESPA SUB BASIN 2 - LIQENAS POINTS OF SAMPLING (POS) AT ALBANIAN PART

Net number	Sector's Sub Basin 2	Depth strata	Coordinates		Date	Time of net setting	Time of net lifting	Time efforts (min)	Sechi D (m)	T (oC)
1	D	0-3 m	40472416	20544935	24.10.2013	1735	630	815	3	16
2	J	0-3 m	40482543	20562989	25.10.2013	1720	620	780	2,8	15,8
3	E	0-3 m	40474006	20550836	24.10.2013	1750	650	780	3	16
4	G	0-3 m	40474930	20551532	26.10.2013	1750	620	790	2,7	15,7
5	P	0-3 m	40495369	20561066	26.10.2013	1800	630	750	2,7	15,7
6	H	0-3 m	40480004	20555836	27.10.2013	1735	605	790	2,5	15,6
7	N	0-3 m	40492328	20562417	27.10.2013	1755	620	785	2,5	15,6
8	A	0-3 m	40462969	20543651	23.10.2013	1740	600	780	3,2	16,1
9	K	0-3 m	40485494	20561996	25.10.2013	1740	640	780	2,8	15,8
10	D	0-3 m	40464469	20543963	23.10.2013	1750	610	780	3,2	16,1
11	H	0-3 m	40472747	20544581	28.10.2013	1750	700	830	2,6	15,5
12	J'	3-6 m	40463016	20545917	23.10.2013	1810	620	730	3,2	16,1
13	E	3-6 m	40472680	20551469	23.10.2013	1825	630	725	3,2	16,1
14	H'	3-6 m	40480005	20561153	24.10.2013	1810	715	785	3	16
15	I'	3-6 m	40480672	20562913	24.10.2013	1830	735	785	3	16
16	J'	3-6 m	40462811	20653604	27.10.2013	1815	645	750	2,5	15,6
17	N	3-6 m	40491181	20564645	25.10.2013	1800	700	780	2,8	15,8
18	B'	3-6 m	40464128	20550742	27.10.2013	1835	705	790	2,5	15,6
19	O'	3-6 m	40500811	20564348	26.10.2013	1815	645	750	2,7	15,7
20	V'	3-6 m	40495517	20563374	26.10.2013	1830	700	790	2,7	15,7
21	O'	3-6 m	40491316	20564614	25.10.2013	1820	720	780	2,8	15,8
22	B'	6-12 m	40463540	20552326	23.10.2013	1840	645	725	3,2	16,1
23	J'	6-12 m	40484356	20571581	27.10.2013	1850	725	795	2,5	15,6
24	I'	6-12 m	40474410	20562670	27.10.2013	1910	745	755	2,5	15,6
25	O'	6-12 m	40484356	20571227	25.10.2013	1850	745	815	2,8	15,8
26	F'	6-12 m	40474941	20562749	24.10.2013	1850	755	785	3	16
27	J'	6-12 m	40483819	20571140	25.10.2013	1910	712	722	2,8	15,8
28	N'	6-12 m	40492602	20570327	28.10.2013	1830	730	780	2,6	15,5
29	O'	6-12 m	40495316	20572189	26.10.2013	1845	720	795	2,7	15,7
30	S	6-12 m	40491589	20572373	26.10.2013	1900	745	765	2,7	15,7
31	I'	6-12 m	40475414	20563362	24.10.2013	1905	810	785	3	16
32	F'	6-12 m	40474408	20560551	23.10.2013	1900	705	725	3,2	16,1

LAKE PRESPA SUB BASINS POINTS OF SAMPLING (POS) AT MACEDONIAN PART

SUB BASIN	NET NUMBER	SAMPLING DATE	STRATA (m)	ACTUAL DEPTH (m)	LONGITUDE	LATITUDE
SB3 ASAMATI	1	11.10.2013	3 - 6	6.0 - 6.0	E 21° 01´ 724	N 40° 59´ 210
	2		6 - 12	7.5 - 8.1	E 20° 01´ 609	N 40° 59´ 155
	3		6 - 12	10.2 - 10.5	E 20° 01´ 609	N 40° 59´ 080
	4		6 - 12	11.9 - 11.5	E 20° 01´ 572	N 40° 59´ 038
	5		6 - 12	11.9 - 11.9	E 20° 02´ 187	N 40° 58´ 840
	6	16.10.2013	6 - 12	15.3 - 12.0	E 21° 00´ 771	N 40° 59´ 141
	7		0 - 3	2.4 - 2.4	E 21° 01´ 149	N 40° 59´ 469
	8		0 - 3	2. 6 - 2.6	E 21° 01´ 194	N 40° 59´ 454
	9		0 - 3	2.6 - 2.6	E 21° 01´ 587	N 40° 59´ 341
	10		0 - 3	2.7 - 2.7	E 21° 01´ 883	N 40° 59´ 296
	11		0 - 3	2.5 - 3.5	E 21° 02´ 116	N 40° 59´ 240
	12		3 - 6	3.7 - 3.7	E 21° 02´ 357	N 40° 59´ 118
	13		3 - 6	3.5 - 3.5	E 21° 02´ 536	N 40° 59´ 075
	14		3 - 6	3. 8 - 3.8	E 21° 02´ 705	N 40° 58´ 891
	15		3 - 6	3.7 - 3.7	E 21° 02´ 772	N 40° 58´ 841
SB4 OTESEVO	16	10.10.2013	0 - 3	1.0 - 1.0	E 20° 54´ 153	N 40° 57´ 504
	17		0 - 3	0.0 - 3.0	E 20° 54´ 119	N 40° 57´ 465
	18		0 - 3	0.0 - 3.0	E 20° 54´ 029	N 40° 57´ 372
	19		3 - 6	3.0 - 6.0	E 20° 54´ 073	N 40° 57´ 387
	20		3 - 6	3.0 - 6.0	E 20° 54´ 031	N 40° 57´ 309
	21	11.10.2013	3 - 6	6.0 - 6.0	E 20° 55´ 076	N 40° 58´ 385
	22		6 - 12	8.0 - 9.0	E 20° 55´ 139	N 40° 58´ 356
	23		6 - 12	9.6 - 9.8	E 20° 55´ 150	N 40° 58´ 301
	24		6 - 12	10.0 - 10.7	E 20° 55´ 153	N 40° 58´ 210
	25		6 - 12	11.5 - 11.9	E 20° 55´ 222	N 40° 58´ 115
	26		0 - 3	0.0 - 3.0	E 20° 55´ 260	N 40° 58´ 085
	27		3 - 6	5.0 - 6.0	E 20° 55´ 251	N 40° 58´ 030
SB5 KONJSKO	28	12.10.2013	0 - 3	2.5 - 2.4	E 20° 59´ 219	N 40° 54´ 945
	29		3 - 6	2.8 - 3.6	E 20° 59´ 140	N 40° 54´ 953
	30		0 - 3	1.5 - 2.5	E 20° 58´ 871	N 40° 54´ 961
	31		6 - 12	7.1 - 8.2	E 20° 58´ 887	N 40° 54´ 995
	32		6 - 12	9.3 - 9.7	E 20° 58´ 932	N 40° 55´ 012
	33		6 - 12	12.1 - 8.3	E 20° 59´ 052	N 40° 55´ 001
	34		3 - 6	6.6 - 6.3	E 20° 59´ 252	N 40° 54´ 990
	35		6 - 12	11.7 - 8. 3	E 20° 59´ 354	N 40° 54´ 982
	36	15.10.2013	0 - 3	1.6 - 1.6	E 20° 59´ 317	N 40° 54´ 936
	37		0 - 3	2.2 - 2.2	E 20° 59´ 439	N 40° 54´ 887
	38		0 - 3	2.6 - 2.6	E 20° 59´ 485	N 40° 54´ 836
	39		3 - 6	4.5 - 6.0	E 20° 59´ 219	N 40° 54´ 945
SB6 CENTRAL PLATE	40	13.10.2013	14 - 16	14.0 - 16.0	E 21° 01´ 816	N 40° 54´ 640
	41		14 - 16	14.0 - 16.0	E 21° 01´ 939	N 40° 54´ 550
	42		14 - 16	14.0 - 16.0	E 21° 02´ 101	N 40° 54´ 427
	43		14 - 16	14.0 - 16.0	E 21° 02´ 281	N 40° 54´ 380
	44		14 - 16	14.0 - 16.0	E 21° 02´ 524	N 40° 54´ 321
	45		14 - 16	14.0 - 16.0	E 21° 02´ 675	N 40° 54´ 328
	46		14 - 16	14.0 - 16.0	E 21° 02´ 890	N 40° 54´ 388
	47		14 - 16	14.0 - 16.0	E 21° 02´ 928	N 40° 54´ 524
	48		14 - 16	14.0 - 16.0	E 21° 02´ 947	N 40° 54´ 692
	49		14 - 16	14.0 - 16.0	E 21° 02´ 855	N 40° 54´ 831
	50	14.10.2013	14 - 16	14.0 - 14.0	E 21° 01´ 780	N 40° 57´ 223
	51		14 - 16	14.0 - 14.0	E 21° 01´ 690	N 40° 57´ 360
	52		14 - 16	14.0 - 14.0	E 21° 01´ 580	N 40° 57´ 420
	53		14 - 16	14.0 - 14.0	E 21° 01´ 446	N 40° 57´ 522
	54		14 - 16	14.0 - 14.0	E 21° 01´ 137	N 40° 57´ 605
	55		14 - 16	14.0 - 14.0	E 21° 00´ 923	N 40° 57´ 655
	56		14 - 16	14.0 - 14.0	E 21° 01´ 640	N 40° 57´ 705
	57		14 - 16	14.0 - 14.0	E 21° 00´ 394	N 40° 57´ 727
	58		14 - 16	14.0 - 14.0	E 21° 00´ 206	N 40° 57´ 665
	59	15.10.2013	14 - 16	14.0 - 14.0	E 21° 02´ 401	N 40° 56´ 239
	60		14 - 16	14.9 - 14.9	E 21° 02´ 620	N 40° 56´ 355
	61		14 - 16	14.8 - 14.8	E 21° 02´ 540	N 40° 56´ 521
	62		14 - 16	14.8 - 14.8	E 21° 02´ 435	N 40° 56´ 696
	63		14 - 16	14.8 - 14.8	E 21° 02´ 312	N 40° 56´ 899
	64		14 - 16	14.0 - 14.0	E 21° 02´ 190	N 40° 56´ 960
PELAGIC NET	65	16.10.2013	0 – 6 surface	0.0 - 6.0	E 20° 55´ 495	N 40° 56´ 696

LAKE PRESPA ADDITIONAL SAMPLING DATA (MACEDONIAN PART)

SUB BASIN	SAMPLING DATE	AIR TEMPERATURE (°C)	WATER TEMEPERATURE (°C)	SECHI DEPTH (m)	pH	OXYGEN ($mg \cdot l^{-1}$)	WEATHER CONDITIONS	MOON
SB3 ASAMATI	11.10.2013	15.1	16.5	5.21	8.20	10.35	Setting time clear, overnight rain, lifting time cloudy	First ½ of the moon
SB3 ASAMATI	16.10.2013	15.5	17.0	3.20	8.25	10.25	Setting time clear, overnight rain, lifting time cloudy and rain	½ of the moon
SB4 OTESEVO	10.10.2013	19.4	16.9	2.70	8.31	10.30	Clear	First ½ of the moon
SB4 OTESEVO	11.10.2013	15.5	16.5	2.95	8.20	10.25	Clear	First ½ of the moon
SB5 KONJSKO	12.10.2013	18.0	16.7	3.20	8.25	10.20	Clear, calm	First ½ of the moon
SB5 KONJSKO	15.10.2013	15.1	17.3	3.60	8.33	11.50	Setting time clear, overnight partly cloudy, lifting time cloudy	½ of the moon
SB6 CENTRAL PLATE	13.10.2013	19.0	17.0	3.30	8.32	10.20	Partly cloudy	½ of the moon
SB6 CENTRAL PLATE	15.10.2013	15.9	17.0	3.10	8.57	10.40	Cloudy	½ of the moon

REPORT

MICROBIOLOGICAL INVESTIGATION OF THE WATER OF LAKE OHRID AND RIVER SATESKA, RIVER KOSELSKA AND RIVER CHERAVA

Trans-boundary Monitoring program

Prepared by Dr. Lence Lokoska
Hydrobiological Institute, Ohrid, R. Macedonia

June, 2014

Contents

List of abbreviations

ALPA - Andrade Lactose Peptone Aqua

APHA – American Public Health Association

CCA - Chromocult Coliform Agar

CFU – Colony Forming Units

Ch1 - River Cherava inlet

Kos1 - River Koselska inlet

MPN - Most Probable Number

Sat1 - River Sateska before redirection

Sat 2 - River Sateska middle course

Sat 3 - River Sateska inlet

TC - Total Coliforms

TFC - Total faecal coliforms

WHO – World Health Organization

1. Introduction

The examination of microbiological water quality is obligatory for use-related aspects as such for drinking water, irrigation or recreation. Although microbiological parameters are not standard parameters according to the requirements of the Water Framework Directive (WFD) they are useful to assess the impact especially of pollution causes by domestic wastewater and other organic pollution from diffuse sources. Furthermore analysis of microbiological standard parameters (e.g. fecal indicator bacteria) is obligatory according to the EC-Surface & Drinking Water Directive and the EC-Bathing Water Directive.

Even when biological and chemical water quality is acceptable, microbiological parameters can indicate serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation (Kavka et al. 2002).

The estimation of bacteriological quality of surface waters based on classical sanitary indicators (total coliforms, faecal coliforms, fecal streptococci) may not reflect their safety for the health of bathing people and/or using water for drinking and household purposes. Numerous human diseases having bath in rivers, lakes, pond and coastal sea waters in the area of river and sewage inflow, swimming, swimming pools are associated with the presence of opportunistic pathogens from *Pseudomonas, Aeromonas, Staphylococcus* and other microorganisms groups, being able to generate infections by contact with skin, mucous membrane, nasopharyngeal cavity, respiratory ducts, eyes, ears and urogenital passages. Pyogenic infection of injuries, meningitis, urinary system, respiratory system, inflammation of the middle ear and eyes are typical diseases caused by polluted water where Pseudomonas are found (Pellet et al. 1983; Niewolak and Opieka 2000). Coliforms are therefore now being supplanted by more specific indicators. These include *Escherichia coli* and enterococci which are good indicators of gastrointestinal disease, and Pseudomonas which correlates well with ear and skin infections (Warrington and Jonson 2001).

Wound infections, peritonitis, meningitis, endocarditis, septicemia, corneal ulcers, nosocomial infections, urinary tract infections, gastroenteritis of people who bathe and/or use water in other ways are caused by Aeromonas (Kersters et al. 1996, Kuhn et al. 1997). Aeromonas is aquatic, fresh water bacteria that were identified during the last century as responsible of infectious processes in aquatic animals: amphibians, reptiles, fish, snails and others (EPA Office of Water) 2006. Aeromonas numbers range between 1000–10000 bact./100ml normally in waters not affected by sewage pollution. They can occur in numbers up to 100000 bacteria in fresh water affected by sewage pollution. However standards have been established in Europe for numbers of Aeromonas in drinking water:

- Water after leaving the treatment plant should have no more than 20 bact/100ml
- In the distribution system up to 200 bact./100ml is acceptable.

The purpose of these investigations was to establish the water status of Lake Ohrid and its tributaries (River Sateska, River Koselska and River Cherava according to microbiological aspects (water faecal pollution and water contamination by organic matter) and to assess the impact of the waste water discharges.

To facilitate the interpretation of microbiological water quality data the results were classified according to EC-Bathing water quality directive 2006/7/EEC considering actual expert proposals (Kavka &Poetch, 2008).

2. Parameter and sampling points

Parameters of sanitary aspects of the conditions of the water (coliform bacteria, fecal indicators) for determination of its hygiene status: Total Coliforms bacteria (TC), Total Faecal Coliforms (TFC), Enterococci, Escherichia coli, Pseudomonas, Clostridium perfringens, Aeromonas was investigated.

Furthermore bacteria indicatory of water contamination by organic matter, heterotrophic bacteria was investigated (Colony count 22^{0}C - Aerobic psihrophilic bacteria and Colony count 37^{0}C -Aerobic mesophilic bacteria).

Water samples were collected from following sampling sites on the lake:

MK I Kalishta (N 41o09'025 E 20o39'078),

MK II Grashnica (N 41o07'043 E 20o47'260),

MK III Velidab (N 40o59'237 E 20o47'965),

MK IV St. Naum (N 40o54'855 E 20o44'499),

MK V Pelagic zone (N 41o03'730 E 20o45'000)

Additionally water samples were collected from the following sampling sites at the main tributaries:

Sat 1 River Sateska before redirection (N 41o13'569 E 20o44'755),

Sat 2 River Sateska middle course (N 41o12'959 E 20o44'811),

Sat 3 River Sateska inlet (N41o10'071 E 20o43'620),

Kos 1 River Koselska (N41o07'355 E 20o46'329),

Che 1 River Cerava inlet (N 40o55'402 E 20o45'376)

Figure 1. Sampling site Lake Ohrid and tributaries

3. Description of investigations

A number of documents contain detailed information on taking samples for analysis of microbiological parameters and their storage and transportation (ISO standard, WHO 2001, 2003, Standard methods -AFA-AWA-WPCF, 2005).

At the water surface, samples for microbiological analyses were collected directly into sterile glass bottles. In deeper water layer, a 2,5 l Ruttner sampler was used.

Collecting of samples was done by bout (from lake – one day) and by car from tributaries (River Sateska, Koselska and Cherava – another day).

Samples were collected seasonally from spring 2013 to winter 2014, in four campaigns at eight different stations:

- first campaign in spring
  11.06.2013, littoral and tributaries when the weather was cloudy
- with slight wind;
  13.06.2013, pelagial when the weather was cloudy with slight wind
- second campaign in summer
  22.07.2013, littoral and tributaries when the weather was sunny;
  24.07.2013, pelagial when the weather was sunny
- third campaign in autumn
  21.10.2013, littoral and tributaries when the weather was sunny, with slight south-west wind;
  24.10.2013, pelagial when the weather was sunny
- fourth campaign in winter 22.01.2014
  littoral and tributaries when the weather was cloudy with north wind
  24.01.2014, pelagial when the weather was cloudy with slight
  north wind

4. Transport and storage of samples

The water samples for microbiological analyses were stored in 500 ml sterile glass bottle, and transported to the laboratory in the hand refrigerator at low temperature, and analysed within a maximum of 18-24 h after collection.

5. Analysis

Microbiological indicators of sanitary quality and indicators for organic assessment of contamination were analyzed using standard procedures (Official Gazette of R. Macedonia, number 18/99; Official Gazette of R. Macedonia, number 46/2008); EU-Bathing Water Directive 2006/7/EC and repealing directive 76/160/EEC, EU-Surface & Drinking Water Directive 75/440/EEC, ISO standard, WHO, Standard methods (AFA-AWA-WPCF, 2005).

The following microbiological indicators for Drinking water quality and Bathing water quality ware analyzed: Total Coliforms (TC), Total faecal coliforms (TFC), Enterococci, *Eschericha coli*, Pseudomonas, *Clostridium perfringens*, Aeromonas and heterotrophic bacteria (colony count 22 and 37^{0}C).

Generally *E. coli* and coliforms are used as indicators of contamination of waters, but recent research has shown that Aeromonas are equally important as a measure of water quality because they are implicated in a range of diseases of animals and humans. Pseudomonas correlates with ear and skin infections (APHA-AWA-WPCF, 2005).

The methods for microbiological analyses are according to Standard Methods (for the examination of water & waste water APHA-21st Edition, 2005) and according to ISO standards, Rodina (1971), Kohl (1975), Kavka (1994).

Bacteriological analyses

The heterotrophic plate count includes all of the microorganisms that are capable of growing in a nutrient-rich solid agar medium:

- Total count (CFU/1ml or bact/ml) of heterotrophic bacteria (psychrophilic) were determined on after 5-7 days incubation at 22⁰C, in plate count nutrient agar,
- Total count (CFU/1ml) of heterotrophic bacteria (mesophilic) - in plate count nutrient agar after 72 h incubation at 37⁰C.

Total coliforms (TC) and *E. coli* were determined according membrane filter method, using Chromocult Coliform agar (CCA, Merck) after incubation 24 h on 37⁰C. All blue colonies which developed on CCA were accounted as faecal coliforms and *E. coli* and with red are accounted as total coliforms (Farnleitner et al. 2001). This method is approved and accepted as alternative method according to EU water directive and compared to the reference method of the ISO9308-1.

Total faecal coliforms (TFC) were determined according membrane filter method, using Chromocult Coliform agar (CCA, Merck) after incubation 24 h on 44⁰C, and Multiple-Tube Fermentation procedure (now referred to as the Most Probable Number or MPN procedure on ALPA (Andrade Lactose Peptone Aqua). The most probable number of colifornn bacteria (MPN) was determined by fermentation test on ALPA - liquid medium. The test tubes with observed change of color and presence of gas in the Durham tubes were evidenced as positive (APHA-AWWA-WPCF, 2005).

Enterococci were cultivated on Bile esculin agar (membrane filter technique). Cultural characteristic, after 18-24h at 37⁰C with brown colonies and luxuriant growth.

For Pseudomonas enumeration 100 ml water was filtered through sterile membrane filter. Each membrane filter was placed on a plate with Pseudomonas (Cetrimide) agar after 72h incubation at 37⁰C. *Pseudomonas aeroginosa* colonies are fluorescent yellow-green.

Selective Aeromonas medium was used for isolation and enumeration, with membrane filtration and plate method after 24h incubation at 37⁰C.

Clostridium perfringens was determined on the Wilson-Blair medium in pasteurised (80⁰C/10min) water samples, after 18h incubation at 37⁰C. *Cl. Perfringens* produces black colonies.

Classification was performed with class limit values shown in Table 1, proposed by Kavka and Poetsch (2002), and used in the Joint Danube Survey (Kavka et al., 2006).

Table 1: Class limit values for microbial pollutions of rivers assessed by bacteriological standard parameters according to Kohl (1975, modified), Kavka & Poetsch (2002, modified) and in consideration of EU-Bathing water quality directive 2006/7/EEC (target values for E. coli and enterococci concerning a good microbiological bathing water quality = target values in this table) and directive 76/160 EEC (imperative value for total coliforms concerning bathing water quality = target value in this table);

TV=Target Values; FS=faecal streptococci

a) faecal pollution,

Classification of faecal pollution		CLASS				
		I	II (TV)	III	IV	V
Parameter	Faecal pollution	little	moderately	critical	strongly	excessively
Escherichia coli (Fecal coliforms)	In 100 ml water	≤ 100	> 100 -1 000	> 1 000 -10 000	> 10 000 -100 000	>100 000
Intestinal Enterococci (FS)	In 100 ml water	≤ 40	> 40 -400	> 400 -4 000	> 4 000 -40 000	>40 000
Total Coliforms	In 100 ml water	≤ 500	> 500 -10 000	> 10 000 -100 000	> 100 000 -1000 000	>1000 000

b) pollution by easily degradable organic matter

Classification of organic pollution		CLASS				
		I	II	III	IV	V
Parameter	Organic pollution	little	moderately	critical	strongly	excessively
Heterotrophic Plate Count 22 ^{0}C	In 1 ml water	≤ 500	> 500 -10 000	> 10 000 -100 000	> 100 000 -750 000	>750 000

6. Evaluation

6.1. Total coliform bacteria

The term total coliform bacteria refers to a number of bacteria including Escherichia, Klebsiella, Citobacter and Enterobacter. They are associated with feces of warm-blooded animals and are also present in soil. Total coliforms are not specific indicators of faecal pollution (Ashbolt et al, 2001), but can provide basic information on source water quality.

Figure 2 shows seasonal coliform counts for the different stations. It is evident that littoral water at Grashnica has much higher coliform counts (34 000 bact/100ml) than the investigated part of Lake Ohrid littoral. No coliform bacteria were found in the water of lake Ohrid pelagial. Total coliform bacteria were found in all samples of water from lake tributaries, with maximum in River Cherava (34 500 bact./100ml).

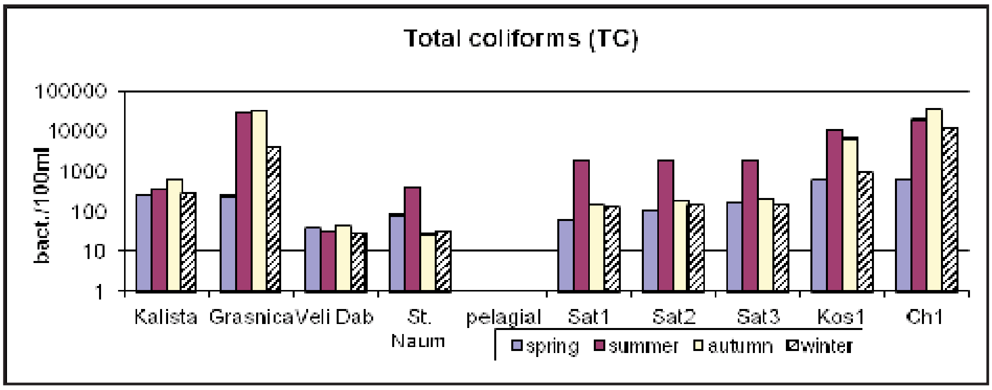

Figure 2: Seasonal fluctuation of total coliforms in the water of Lake Ohrid and its tributaries

According to presence of the total coliforms, Kavka & Poetsch (2002) classifications, the Lake Ohrid is unpolluted in the pelagic point and some part of the littoral except at Kalista in the summer is moderately polluted (II class) and Grashnuca where is critical polluted in summer and autumn (III class) and moderately (II) in winter. Based on this classification the waters of the Lake Ohrid tributaries are in: River Sateska I class, except in summer (II class); River Koselska is moderately (II spring and winter) to critical (III-summer and autumn); River Cherava moderately (II class) in spring and critical (III) in other three seasons.

According to presence of the total coliforms and EU-Bathing water quality directive (1976/160/EEC), Lake Ohrid shows excellent water quality, except littoral zone at Grashnica

6.2. Total fecal coliform bacteria

Fecal coliform bacteria are a subgroup of the total coliform group. They are indicators of fecal contamination because they are restricted to the intestinal tract of warm-blooded animals. Their use enables separation of bacteria from soil and fecal origin.

Total fecal coliform bacteria varied from 17 (at St. Naum – winter) to 28 700 bact./100ml (at Grashnica – autumn). These bacteria are not registered in the littoral water at Velidab (except in autumn) and in pelagic water of Lake Ohrid. Number of total coliform in the water of Lake Ohrid tributaries is ranged from 64 bact./100ml (Sat1- spring) to 1986 bact./100ml (Sat 3 –summer in River Sateska; from 622 to 11 200 bact./100 ml in River Koselaska and from 632 to 34 500 bact./100ml in River Cherava.

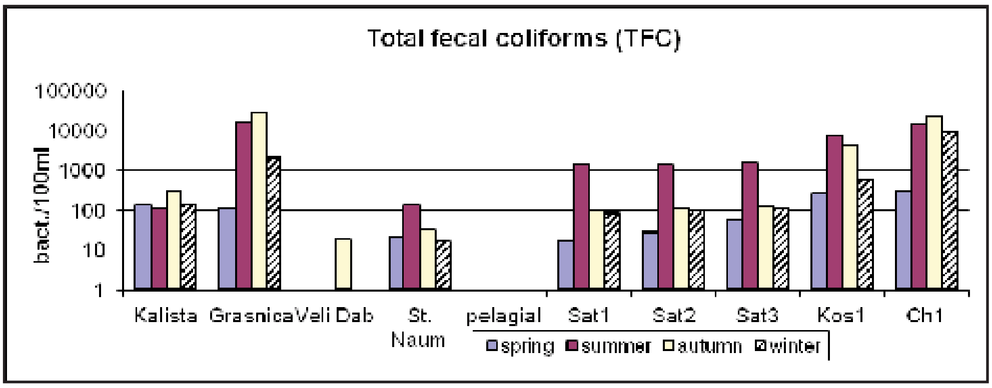

Figure 3: Seasonal fluctuation of total fecal coliforms in the water of Lake Ohrid and its tributaries

6.3. Escherichia coli

Escherichia coli is a type of fecal coliform and is an indicator of fresh fecal pollution It is evident from the results that *Escherichia coli* number varied from 2 bact/100ml at St. Naum (autumn) to 16 800 bact./100ml in the littoral water at Grashnica and from 58 to 19 600 bact./100ml (River Cherava) in the water of tributaries. The presence of these bacteria was not evidenced in the water of lake pelagial and littoral at Velidab (except autumn 4 bact./100ml) during all seasons, and in River Sateska at all tri sampling points during spring.

Escherichia coli occur in high numbers in human and animal feces, sewage and water subject to recent fecal pollution. Water temperatures and nutrient conditions present in aquatic ecosystems are highly likely to support the growth of these organisms.

It is generally accepted that *E. coli* and other coliform bacteria can survive in aquatic environments for at least several weeks, depending on the nutriment availability, pH, and water temperature.

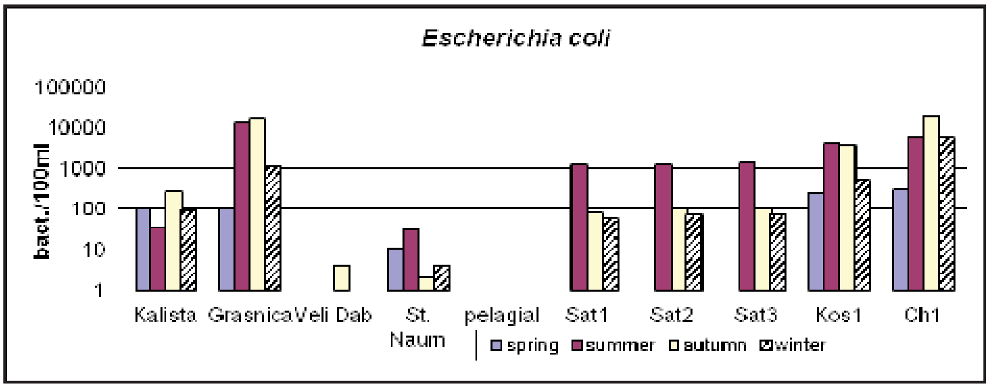

Figure 4: Seasonal fluctuation of Escherichia coli in the water of Lake Ohrid and its tributaries:

For better evaluation of fecal pollution, the number of fecal coliforms *(E.coli)* was also used in investigations. This parameter indicated Ist class water quality in the Lake Ohrid, except the one at Grashnica where the water is moderately polluted in spring (II), strongly polluted in summer and autumn (IV class) and (III) critical in winter (Kavka & Poetsch 2002, EU-Bathing water quality directive 76/160/EEC, 2006/7/EEC); the water of River Sateska is critical fecal polluted (III) et all three localities only in summer; River Koselska is moderately to critical contaminated (II-III) and River Cherava is mainly critical to strongly fecal polluted (III-IV class).

According to the presence and the numbers of *E. coli* and EU-Bathing water quality directive (1976/160/EEC, 2006/7/EEC), Lake Ohrid is with Excellent quality, except reon et Grashnica.

6.4. Enterococci

Enterococcus (Fecal streptococci) is another fecal bacteria and is the most sensitive of the indicator bacteria. Important advantages of this group are that they tend to survive longer in water environments than E. coli(or thermotolerant coliforms), are more resistant to drying and are more resistant to chlorination.

Intestinal enterococci are typically excreted in the faeces of humans and other warm-blooded animals. Some members of the group have also been detected in soil in the absence of faecal contamination. Intestinal enterococci are present in large numbers in sewage and water environments polluted by sewage or wastes from humans and animals.

The tributaries contain much more enterococci in comparison with the littoral region, while in the pelagic region enterococci were not registered. Highest enterococci concentrations were also observed in the littoral water at Grashnica (12 900 bact./100ml) and in the water of River Cherava (19 400 bact./100ml) during summer season.

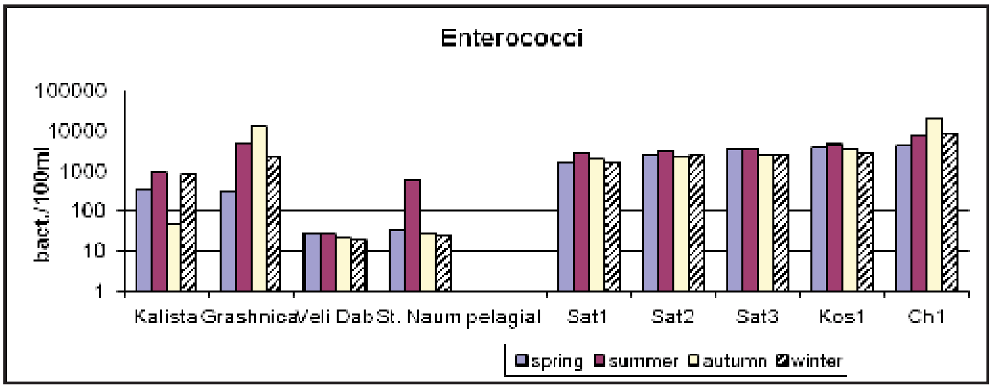

Figure 5: Seasonal fluctuation of enterococci in the water of Lake Ohrid and its tributaries

The number of enteroccoci detected in Lake Ohrid shows that pelagial water is mainly unpolluted, little contamination at Velidab and St.Naum except in summer (critical); moderate to critical (II, III) at Kalishta and mainly strongly fecal contamination at Grashnica (IV); River Sateska is moderately polluted (II class) in all seasons; River Koselska is mainly critical (III class) and River Cherava is excessively polluted (V class0) for all four seasons.

According to the presence and the numbers of enterococci, and EU-Bathing water quality directive (1976/160/EEC, 2006/7/EEC), Lake Ohrid shows excellent quality (pelagial and Veli Dab) except at St. Naum in summer and Kalishta and Grashnica in all seasons.

6.5. Pseudomonas

Pseudomonas aeruginosa is a common environmental organism and can be found in faeces, soil, water and sewage. It can multiply in water environments and also on the surface of suitable organic materials in contact with water.

Pseudomonas was not identified in the water of Lake Ohrid pelagial during the all four seasons, and in the littoral water at Velidab (except in spring). In the littoral water at St Naum Pseudomonas was registered only in spring and summer. The highest number of this bacteria characterized locality Grashnica (238 bact./100ml) and River Koselska (298 bact,100ml).

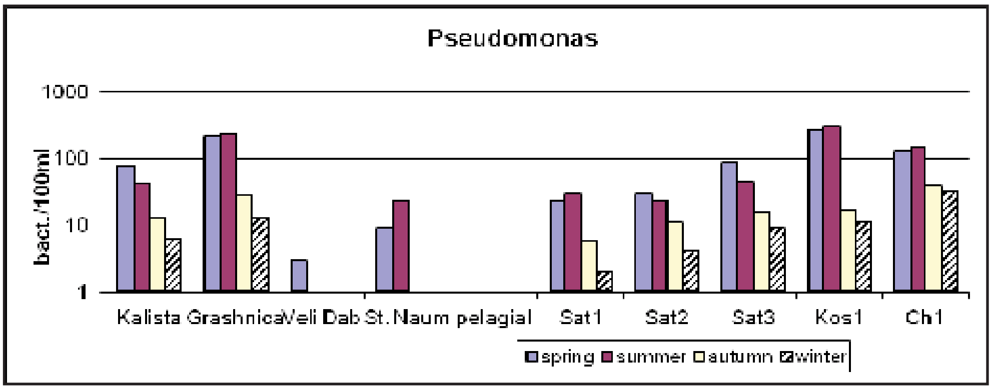

Figure 6: Seasonal fluctuation of Pseudomonas in the water of Lake Ohrid and its tributaries:

6.6. Aeromonas

Generally *E. Coli* and coliforms are used as indicators of contamination of waters. But recent research has shown that Aeromonas are equally important as a measure of water quality because they are implicated in a range of diseases of animals and humans.

Aeromonas was registered in all investigated localities during all four seasons. The highest number of bacteria characterized locality Grashnica (163 200 bact/100ml –summer) and River Cherava (197 000 bact./100ml –summer) and the lowest number was recorded at locality Lake Ohrid Pelagial (2 bact./100ml –spring).

Aeromonas range between 1000–10 000 bact./100ml normally in waters not affected by sewage pollution. They can occur in numbers up to 100 000 bacteria in fresh water affected by sewage pollution.

However standards have been established in Europe for numbers of Aeromonas in drinking water: Water after leaving the treatment plant should have no more than 20 bact/100ml. In the distribution system up to 200 bact./100ml is acceptable.

Aeromonas species are increasingly recognized as enteric pathogens; they possess several virulence factors associated with human disease, and represent a serious public health concern. These findings demonstrate the presence of potentially pathogenic and multi-drug- resistant A. hydrophila in the surface waters, thereby indicating a significant risk to public health.

Aeromonads are ubiquitous in the aquatic environment (Holmes et al., 1996). Aeromonas spp. cause disease in poikilothermic animals, and occasionally in mammals. Diseases caused by aeromonads represent a significant source of loss to the aquaculture industry.

Aeromonas species are autochthonous (natural inhabitants) to the aqueous environment where they may be pathogenic for poikilotherms (cold-blooded animals).

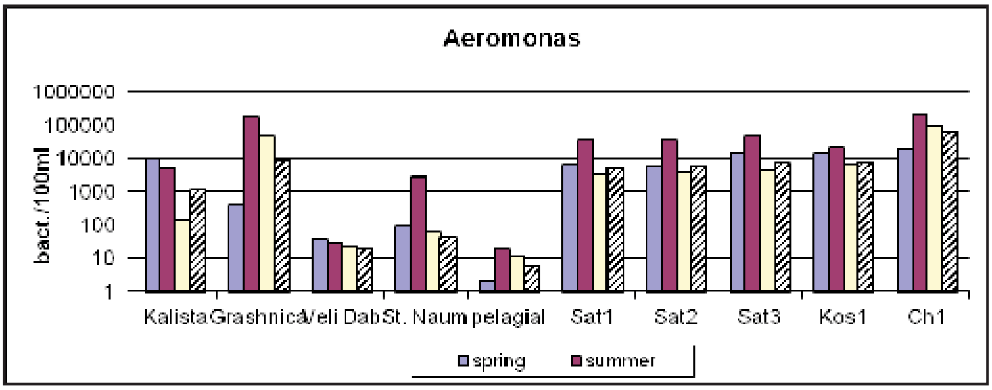

Figure 7: Seasonal fluctuation of Aeromonas in the water of Lake Ohrid and its tributaries

Continuous monitoring of surface waters is important to identify potential water-borne pathogens and to reduce the health risk caused by the genus Aeromonas.

6.7. Clostridium Perfringens

Cl. perfringens, is a member of the normal intestinal flora of 13–35% of humans and other warm-blooded animals. Other species are not exclusively of faecal origin. Like *E. coli, Cl. perfringens* does not multiply in most water environments and is a highly specific indicator of faecal pollution.

Cl. Perfringens was not found in the water of Lake Ohrid pelagial, in the littoral water at Velidab, and St. Naum only in spring and winter. The maximums of this bacteria was registered in the water at Grashnica (4 800bact/100ml), in the River Sateska inlet (Sat 3 – 4 667 bact/100ml) and in the River Cherava (5 800 bact/100ml).

Clostridium perfringens and its spores are virtually always present in sewage. The organism does not multiply in water environments. *Clostridium perfringensis* present more often and in higher numbers in the faeces of some animals, such as dogs, than in the faeces of humans and less often in the faeces of many other warm-blooded animals. The numbers excreted in faeces are normally substantially lower than those of *E. coli*.

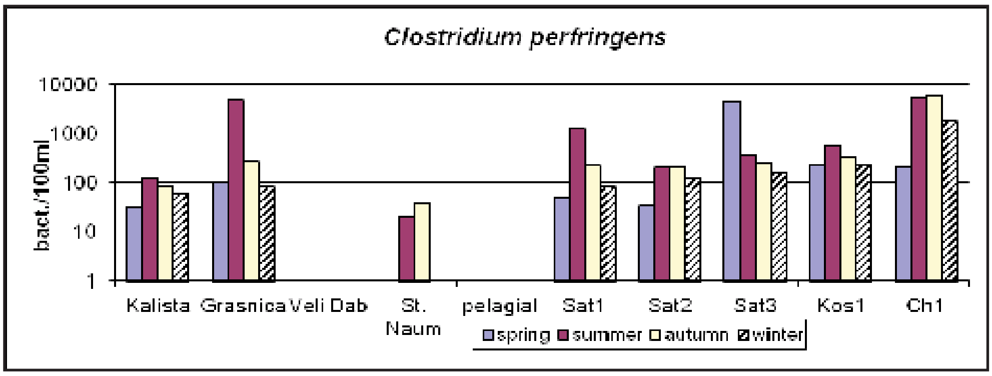

Figure 8: Seasonal fluctuation of Cl. perfringens in the water of Lake Ohrid and its tributaries

6.8. Heterotrophic bacteria (colony count 22 ^{0}C and 37 ^{0}C)

The number of heterotrophic bacteria increases with the trophy and contamination of water bodies with organic matter. Because of the relatively high values of the allohtonous and autochthonous dissolved biodegradable organic matter in littoral zone, there is strong relation between autotrophic and heterotrophic communities, that have significant influence to the whole lake metabolism (Chrost & Overbeck, 1990).

The highest number of heterotrophic bacteria was found in the water of River Cherava (148 800 bact/ml), and in the littoral of the Lake at Grashnica (19 560 bact/ml).

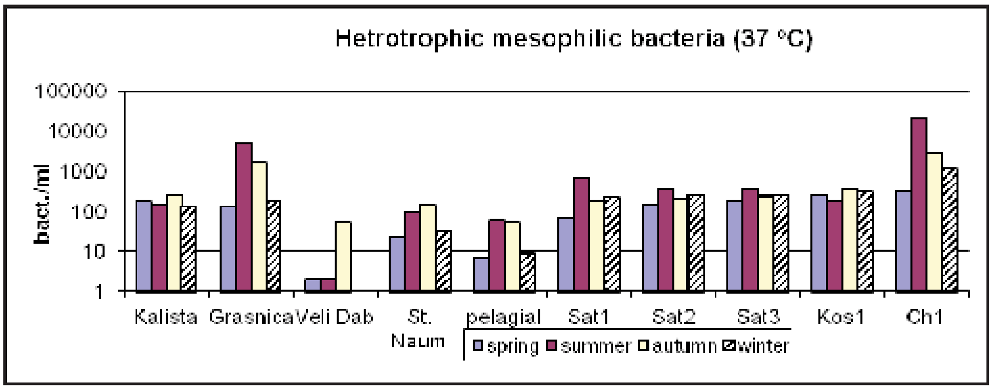

Figure 9: Seasonal fluctuation of Heterotrophic bacteria (370C) in Lake Ohrid and tributaries:

The number of mesophilic bacteria ranged from 0 bact/ml (at Velidab -winter) to 5 184 bact/ml (at Grashnica-summer) in the lake water, and from 68 bact/ml (Sat1) to 21 280 bact/ml (River Cherava) in the water of Lake Ohrid tributaries. Such a high presence of mesophilic heterotrophic bacteria indicates the possibility of eutrophication, above all eutrophication of anthropogenic origin. This situation is proved with the presence of coliform bacteria and fecal indicator bacteria. The high presence of mesophilic bacteria is in correlation with high presence of coliform bacteria.

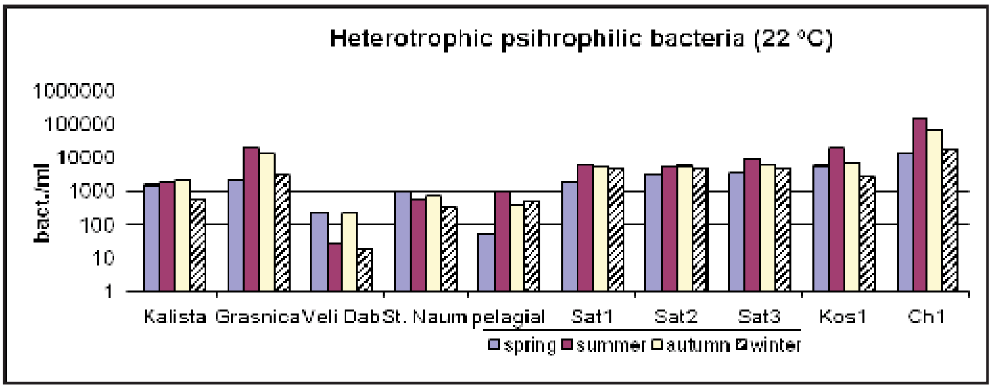

Figure 10: Seasonal fluctuation of Heterotrophic bacteria (220C) in Lake Ohrid and tributaries:

The number of psychrophilic bacteria was between 19 bact/ml (at Velidab -winter) to 19 560 bact/ml (at Grashnica-summer) in the lake water, and from 1 890 bact/ml (Sat1) to 148 800 bact/ml (River Cherava -summer) in the water of Lake Ohrid tributaries.

A smaller number of mesophilic bacteria then psychrophilic heterotrophic bacteria has been observed.

The number of heterotrophic bacteria increases with the trophy and contamination of water bodies.

The water quality according to the psychrophilic heterotrophic bacteria Lake Ohrid pelagial, littoral at Velidab is in I class, at St Naum mostly in II class (except winter- I). In another localities number of this bacteria corresponded with moderate, critical to strongly organic pollution (classes II, III, IV): at Kalishta is in II class, and at Grashnica is in the range of II to III class; River Sateska is in II class at all three localities, River Koselska mainly is II and III in summer and River Cherava is mainly in III and IV class in summer (Kohl, 1975).

7. Classification

Potential sources of bacterial contamination include surface water runoff, animal waste, and leachate from sewage disposal systems and improperly disposed of human waste from visitors to the lake and rivers.

The interpretation of microbiological water quality data, and the results are classified according Kohl (1975), EU-Bathing water quality directive (1976/160/EEC and 2006/7/EEC) and EU expert proposals -Kavka & Poetsch (2006).

Microbiological determinants indicated a good bacteriological water quality at three sampling sites (pelagial, Veli Dab and St. Naum). At Kalishta heterotrophs and faecal indicator bacteria (*E. coli*, enterococci) reflect moderate organic pollution (class II) and low to moderate faecal contamination of water (class I and II). At Grashnica was recorded critical contamination by faecal bacteria in all seasons, with maximum in summer (class III and IV). The great abundance of bacteria in the reon of Grashnica, is a result of the contribution of the River Velgoshka, which is under a big anthropogenic influence, loaded with nutrients, seriously polluted, with large number of bacteria.

All three sampling stations at River Sateska (before redirection, middle course and inlet) have the same water quality, low variability of all bacterial numbers and slight increase toward inflow. Presence of heterotrophic bacteria indicated moderate pollution by organic matter. Critical concentrations of enterococci were observed in all season (class III) and *E. coli* during in summer period (class III).

The water of River Koselska is slightly polluted by total coliforms, heterotrophs and *E.coli* for all season (class II), except in summer – critical contaminated (class III). This river was critical (class III) to strongly contaminated (class IV in summer) by enterococci.

River Cherava is most polluted. Presence of heterotrophic bacteria indicated critical (class III) to strongly (class IV in summer) pollution by organic matter. According pollution with *Escherichia coli* this river is moderately polluted in spring (class II), critical polluted (class III) in winter and autumn and strongly polluted in summer (class IV). The river water is strongly polluted (class IV) in all seasons according to presence of enterococci.

The results of the investigations show that the water of Lake Ohrid pelagial is very clear (unpolluted) and could be used for drinking without any treatment.

Lake Ohrid has excellent Bathing Water Quality except littoral zone at Grasnica.

The obtained values for faecal coliforms, *E. coli*, enterococci and Pseudomonas in the water of rivers Sateska, Koselska and Cherava, exceed the prescribed values for water quality for irrigation (Ashbolt 2001, WHO 2001) .

Figure 11: Microbiological map of fecal pollution assessed by the concentrations of fecal coliforms and additionally concentrations of enterococci: blue coloured symbols: class I-little fecal polluted; green; class II-moderately; yellow: class III-critica,; orange: class IV-strongly; red: class V –excessively fecal polluted

Before sewer system around Lake Ohrid has been modernized the discharge of untreated domestic waste water had harmful impact to the water quality (Lokoska, 2000). Nutrient loads of the tributaries are still relevant to the eutrophication processes in the Lake, especially in the estuaries.

The region around Grasnica is still affected by wastewater discharges from the River Velgoska. Other significant indicators of this pollution are the gray color of the river water, unpleasant smell, and noticeable waste. The well known, "City Beach" (in the region of Grasnica), once an attractive place to visit, today (and years back) is forbidden for tourists.

8. Recommendations

The tributaries of Lake Ohrid are characterized by very heavy initial organic loads of industrial and municipal wastewaters followed by an abrupt increase in the heterotrophic bacteria and faecal coliform counts. Otherwise in the some part of littoral and in the pelagic region coliforms were not registered. The water of River Cherava is most polluted by fecal and organic contaminants.

Microbiological investigations have shown that, in the water at Kalishta a presence of coliform bacteria and fecal indicator *Escherichia coli* is noticed, which points at an infiltration of fecal waste water, a result of an anthropogenic influence.

The results indicate pollution of the areas near the settlements and tourist localities. Unfortunately the littoral region still is under impact from the sewerage, industrial and drain waters reach in pollutants and nutrients that lead to increased eutrophycation as it is in the Grasnica area. Furthermore numerous touristic objects located near the shore are potential polluters.

Unfortunately, the investigations show dramatic conditions in the littoral, even in the strictly protected areas as St Naum Springs. There are numerous tourist sites located in this region, that have "septic thanks" of which more of them have obsolete and non-functional treatment systems (s.c. putox), so there is frequent overflowing of the sewerage directly or through the tributaries flow in the lake. This situation requires permanent control and maintenance.

Continuous and complex investigations of the littoral region are necessary, especially most loaded areas, because of pollution control, hold or minimizing, aiming of protection of the good ecological status of Lake Ohrid at last.

Microbiological analyses must be included in all investigations performed on Lake Ohrid and other aquatic ecosystems, as a good indicator for water quality, water categorization and determination the source of pollution.

Even when biological and chemical water quality is acceptable, microbiological parameters can indicate serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation (Kavka et al. 2006).

Recommendations for future microbiological monitoring:

- Microbiological water quality was described by one proposed assessment method and supported the detection of anthropogenic impacts;
- Microbiological results support ecological and physico-chemical assessment of the natural state and water quality of the Lake Ohrid and its tributaries;
- Microorganisms are very sensitive indicators for the detection of fecal and organic pollution caused by raw sewage, municipal waste water treatment plants and diffuse impact from farmland; quantitative results from indicator bacteria facilitate the detection of hot spots;
- Microbiological investigations are obligatory for testing compliance with the requirements for the utilizations of water for drinking, bathing, irrigation and another economic purpose.

9. Summary

Bacteria are ideal sensors for the indication of microbial pollution of surface water ecosystems because of their fast response to changing environmental conditions. Even when biological and chemical water quality is acceptable, microbiological parameters can indicate immediately serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation.

During all four sampling campaigns on Lake Ohrid and its tributaries (Sateska, Koselska Cherava) the obtained results for investigated bacteria showed that generally, their values in the rivers and some part of littoral are with a great variability, and higher than in the pelagial water of Lake Ohrid.

In the water of Lake Ohrid pelagial coliforms bacteria, faecal indicators and Pseudomonas were not detected.

The lowest bacterial number in all seasons was recorded at locality Veli Dab of the lake littoral because it is located outside of settlements and anthropogenic influence.

Microbiological determinants indicated a good bacteriological water quality at three sampling sites (pelagial, Veli Dab and St. Naum). At Kalishta heterotrophs and faecal indicator bacteria (E. coli, enterococci) reflect moderate organic pollution (class II) and low to moderate faecal contamination of water (class I and II). At Grashnica was recorded critical contamination by faecal bacteria in all seasons, with maximum in summer (class III and IV). The great abundance of bacteria in the reon of Grashnica, is a result of the contribution of the River Velgoshka, which is under a big anthropogenic influence, loaded with nutrients, seriously polluted, with large number of bacteria.

All three sampling stations at River Sateska (before redirection, middle course and inlet) have the same water quality, low variability of all bacterial numbers and slight increase toward inflow. Presence of heterotrophic bacteria indicated moderate pollution by organic matter. Critical concentrations of enterococci were observed in all season (class III) and E. coli during in summer period (class III).

The water of River Koselska inlet was found to be moderately contaminated by total coliforms, heterotrophs and E.coli for all season (class II), except in summer – critical contaminated (class III). This river was critical (class III) to strongly contaminated (class IV in summer) by enterococci.

The results of microbiological investigation showed that water of River Cherava inlet is most polluted. Presence of heterotrophic bacteria indicated critical (class III) to strongly (class IV in summer) pollution by organic matter. According contamination with Escherichia coli this river is moderately polluted in spring (class II), critical polluted (class III) in winter and autumn and strongly polluted in summer (class IV). The river water is strongly polluted (class IV) in all seasons according to presence of enterococci.

During the all sampling campaign, the highest abundance of Pseudomonas was recorded in the River Koselska (298bact./100ml), and in the water at Grashnica (238bact./100ml) during summer period. The lowest number of Aeromonas (2 bact./100ml) was evidenced in the water of Lake Ohrid pelagial. Its maximal representation was in the water of River Cherava (197000 bact/100ml) and in the water at Grashnica (163200 bact./100ml) during the summer period, show that these waters are affected by sewage pollution.

The results of the investigations show that the water of Lake Ohrid pelagial is very clear (unpolluted) and can be used for drinking without any treatment.

Lake Ohrid has excellent Bathing Water Quality except littoral at Grasnica.

The obtained values for faecal coliforms, E. coli, enterococci and Pseudomonas in the water of rivers Sateska, Koselska and Cherava, exceed the prescribed values for water quality for irrigation.

The region at Grasnica is under the strong influence of wastewaters entered by River Velgoska. The pelagic zone is still oligotrophic balanced as a result of the ability for self-purification; it is still not under the influence of communal, industrial and waste waters of the cost rich of pollutants and nutrients which can result in rapid eutrophication of some parts (low-level) of the littoral zone, such as the areas before the river's delta and settlements.

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Zooplankton of Lake Prespa

Report

January, 2014

Authors

Dr. Goce Kostoski

Dr. Dafina Guseska

Dr. Orhideja Tasevska

Contents

1. Introduction

The members of the zooplankton community are a significant component in the food chain of the lake's ecosystems. As primary consumers in the food chain they greatly participate in the secondary production of continental waters.

The abundance of the zooplankton in fresh waters continuously changes in time and space, thence, the number of individuals, their individual weight and time of development are variable.

Analyzing the zooplankton in the pelagial aquatic ecosystems plays an important role in determining the water quality and the stability of the water ecosystem. Due to their large density, shorter life span, drifting nature, high group/species diversity, and different tolerance to the stress, zooplankter are being used as the indicator organisms for the physical, chemical and biological processes in the aquatic ecosystem.

In the last several decades the Lake Prespa, one of the few ancient, long-lived lakes on earth has been exposed to a great pressure from anthropogenic influences. The lake area has declined significantly and the fields have spread which caused the reduction of the lake littoral zone. All of this causes negative changes, not only to the quality of the water in the littoral area, but to the lake water in general and has a negative influence on the life communities, both benthic and pelagic.

The zooplankton analyses are very important for the lake economy in general, because the Lake Prespa zooplankton, especially Cladocera and Copepoda representatives, are an important food source for some fish species (Popovska-Stanković 1972, 1975; Ocevski et al. 1975).

In the last decade the analyses imply significant alterations in zooplankton, not only in terms of its qualitative, but also its quantitative composition, this, in its part contributes to the structural modifications of the zooplankton components Copepoda, Cladeocera, Rotifera.

The aim of these analyses was to obtain information about the zooplankton and the rate of the changes in Lake Prespa in the last decade as a consequence of the anthropogenic influence and the water level decline.

2. Parameter and sampling points

Parameters in the pelagic zone: Qualitative and quantitative composition of the zooplankton; Index of saprobity

Parameters in the littoral zone: Composition and dynamic of the invertebrate benthic fauna (microzoobenthos: Rotifera and macrozoobenthos: Copepoda, Cladocera); Index of saprobity, Shannon Wiener Diversity Index (H)

Four depths were chosen for the zooplankton analyses (surface, 5 m, 10 m and the deepest layer) in the pelagic zone of Lake Prespa (pelagic point) and 4 localities in the littoral zone of the Lake Prespa (Macedonian part): Ezerani, Ezerani NW, Ezerani NE and Otesevo (Figure 1).

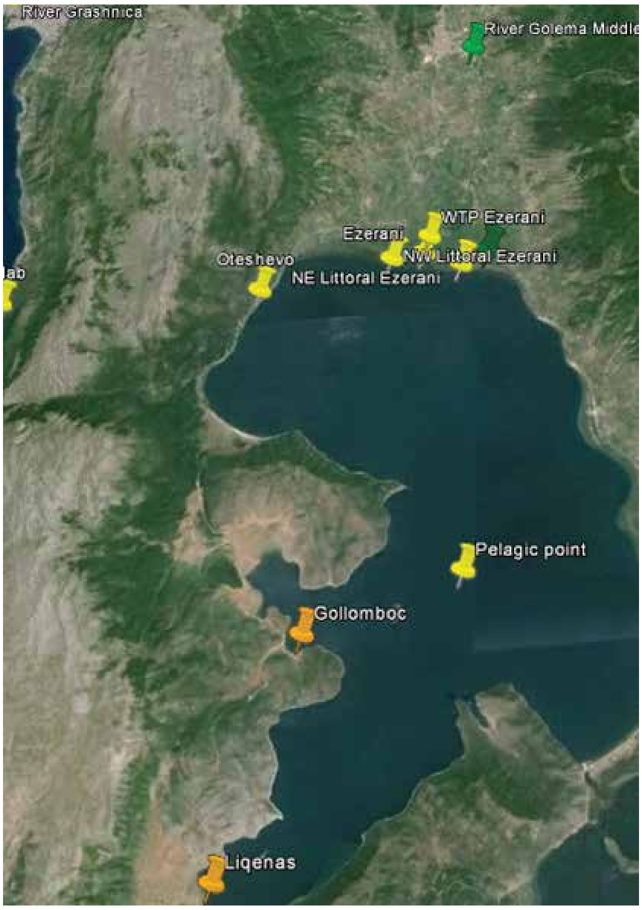

Figure 1. Sampling site Lake Prespa

3. Description of the analyses

The samples for zooplankton analyses were collected on 25.04.2013 in the pelagic zone of Lake Prespa using a Ruttner sampler at four different depths (surface, 5 m, 10 m and 15 m). The material was filtered *in situ* through a sieve (55 µm mesh-size) and preserved with 4% formaldehyde.

The samples in the littoral part of the Lake Prespa were collected on 25.04.2013 using a dredge sampler (with horizontal hauls on the bottom) in four different localities:

- Ezerani – close to the reed beds, on 2 m of depth, where the bottom is of very fine sand;
- Ezerani NW – in the reed beds, on 1.95 m of depth, where the bottom is of fine sand and mollusc shells;
- Ezerani NE – in the reed beds, on 1.75 m of depth, where the bottom is of very fine sand;
- Otesevo – on 2 m of depth, where the bottom is of sand, mud and mollusc shells;

The collected material was filtered *in situ* through a sieve (45 µm mesh-size for Rotifera and 55 µm mesh-size for Copepoda and Cladocera).

4. Transport and storage of samples

The material was stored in 250 ml plastic bottles, and transported to the Department of zooplankton in Hydrobiological institute, Ohrid.

5. Analysis

The material from the pelagic zone was counted with an inverted microscope Leica DMIRB in 50 ml Utermöhl chambers. The qualitative and quantitative composition of zooplankton was analysed and calculated using the saprobic index according to Pantle and Buck (1955) in order to estimate the water quality in this part of the lake.

The material from the littoral zone was analysed with a system microscope Olympus BX43. Identification of Rotifera species was made according to identification guides Kutikova (1970), Koste (1978), Segers (1995). Identification of Copepoda and Cladocera species was made according to identification guides Borutsky (1960), Manuilova (1964), Mazepova (1978).

On the basis of the relative abundance (h) of the identified species and their saprobic indices (s), Index of saprobity (S) was calculated according to Pantle and Buck (1955) and Sladecek (1983).

Species richness i.e. the total number of species, and biodiversity, i.e. the Shannon Wiener Index (H) were determined for each taxonomic group (cladocerans, copepods and rotifers) in all localities.

6. Evaluation

6.1. Pelagic zone of Lake Prespa

In the course of the investigation period 9 zooplankton taxa have been found in Lake Prespa pelagial, 4 of which belonging to Rotifera, 2 to Cladocera, 2 to Copepoda and 1 taxon to Mollusca:

Rotifera

Filinia longiseta (Ehrenberg, 1834)

Kellicottia longispina (Kellicott, 1879)

Keratella cochlearis (Gosse, 1851)

Trichocerca capucina (Wierz. et Zach.,1893)

Cladocera

Bosmina longirostris O.F.Müller, 1785

Daphnia cuculata G. O. Sars, 1862

Copepoda

Arctodiatomus steindachneri Richard, 1897

Mesocyclops leuckarti Claus, 1857

Mollusc

Dreissena polymorpha (Pallas, 1771) (larvae)

The species found are with wide dispersion area, some of them cosmopolitans. Only the species *Arctodiaptomus steindachneri* is endemic for the west part of the Balkan with a limited dispersion area in Lake Ohrid, Prespa and Janina (Kiefer et al. 1978).

In previous analyses (Guseska et al. 2012), during the whole year 18 zooplankton taxa have been found in Lake Prespa pelagial, 10 of which belong to Rotifera, 5 to Cladocera, 2 to Copepoda and 1 taxon to Mollusca.

The results of the quantitative investigations are presented in Figures 2–5.

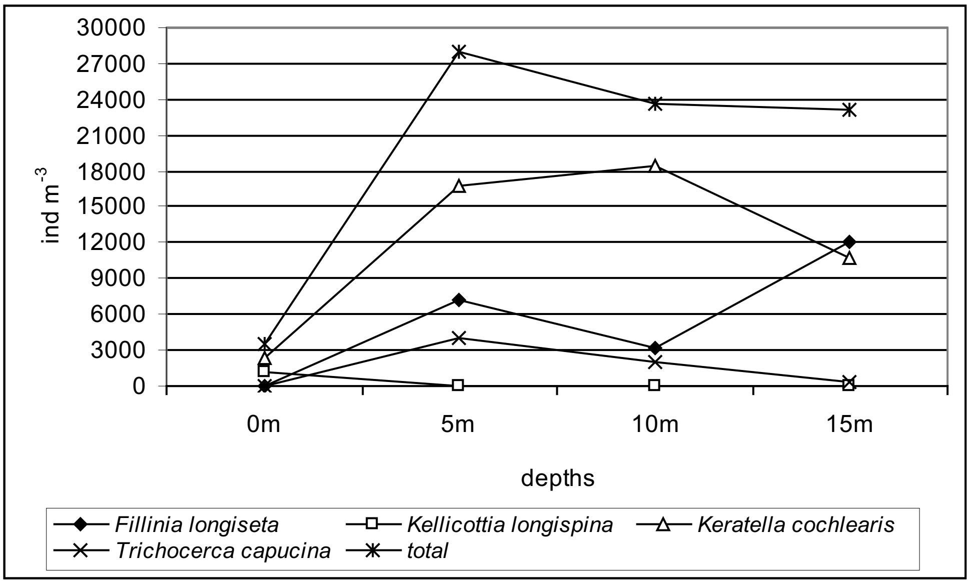

Fig. 2. Vertical distribution of Rotifera's density in Lake Prespa pelagic zone, April, 2013

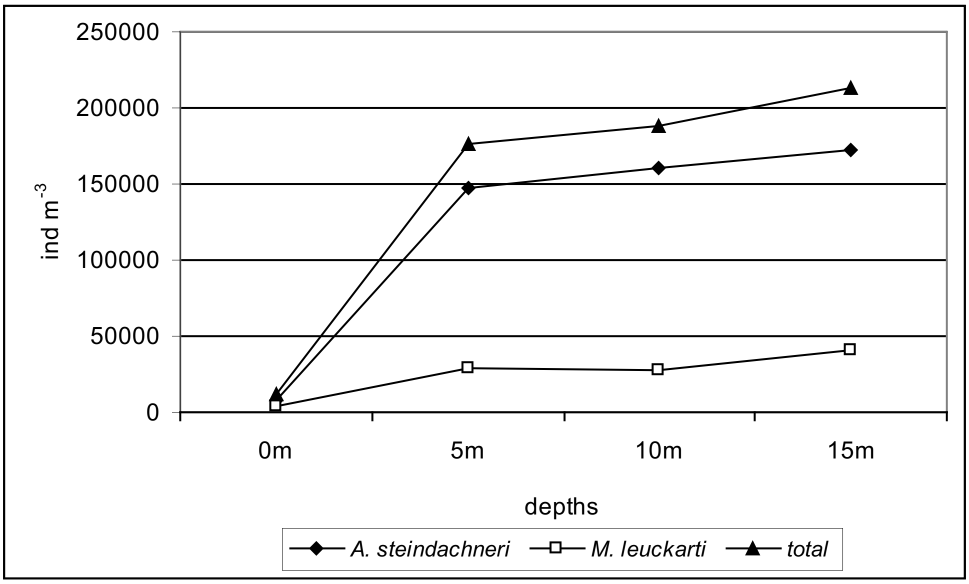

Fig. 3. Vertical distribution of Copepoda's density in Lake Prespa pelagic zone, April, 2013

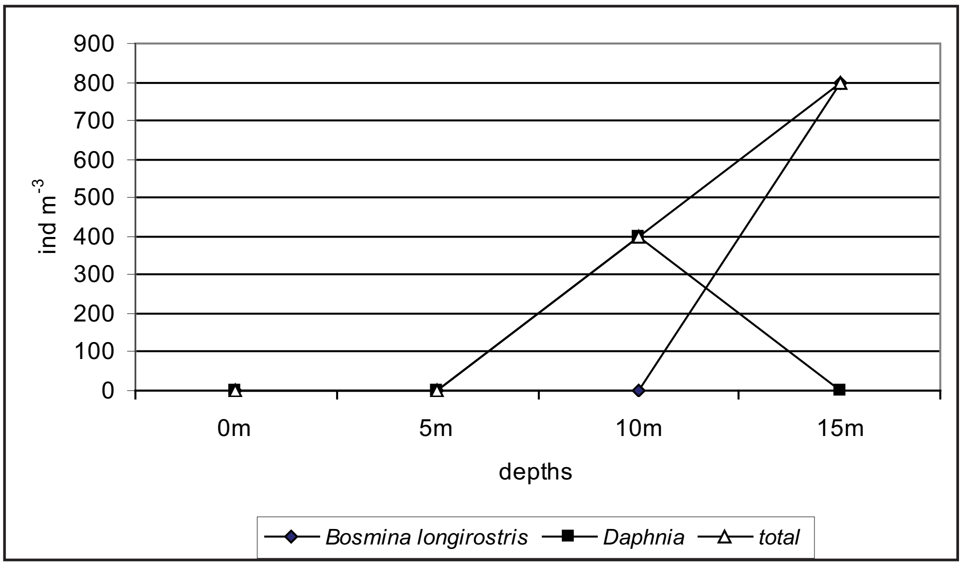

Fig. 4. Vertical distribution of Cladocera's density in Lake Prespa pelagic zone, April, 2013

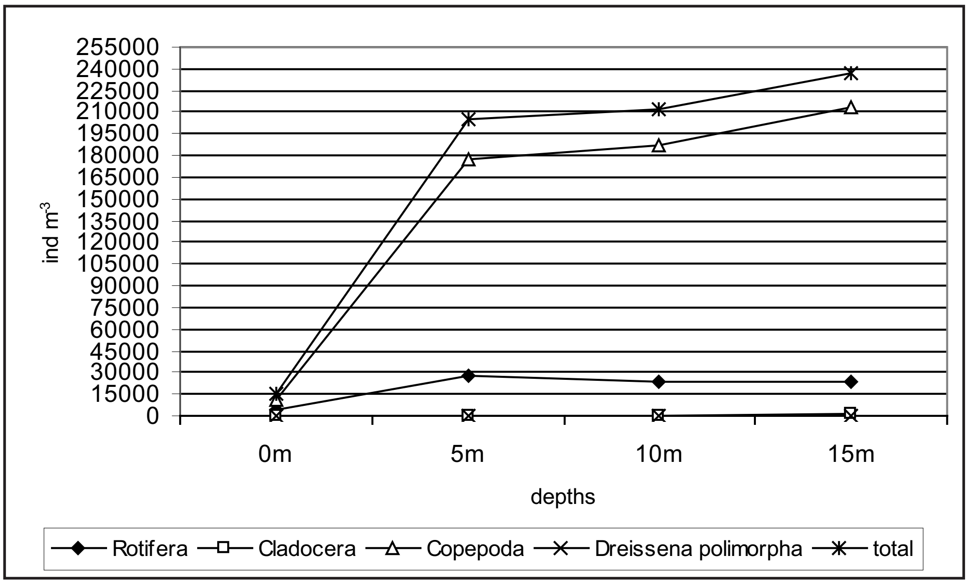

Fig. 5. Vertical distribution of zooplankton density in Lake Prespa pelagic zone, April, 2013

The results showed the spring zooplankton assemblage in the Lake Prespa pelagic region. In this period (April, 2013), the total zooplankton reached higher values in comparison with the spring period 2008–2009 (Guseska et al. 2012).

The total zooplankton population is the most dense at 5 to 10 m of depth. This spatial distribution is conditioned by the abiotic and biotic factors and their interactions in the lake. According to Patceva (2005), the average phytoplankton density reaches its maximum value at the surface layers and at 5 m and 10 m of depth.

According to Hutchinson (1967), competitiveness is the key factor regarding spatial and temporal distribution of various planktonic organisms.

During the analysed period, according to the quantity, the Copepoda representatives were dominant, especially calanoid *Arctodiaptomus steindachneri* and the larval nauplial and copepodid stages which are presented with great numerical values. These filter feeder species increase in numbers in the same time with the phytoplankton peak (Patceva and Mitic 2008).

The cladocerans in this period were presented with lower numerical values. The Cladocera representatives, present in the plankton during the whole year, also play an important part regarding the high maximums of the total zooplankton in late spring and summer. According to the latest investigations (Guseska et al. 2008; Guseska et al. 2012), the numerical values of Cladocera are relatively higher than in the previous investigation periods (Serafimova-Hadzisce 1954, 1958; Sumka 1994; Stankovic et al. 2003).

Fluctuations in the quantity of Copepoda and Cladocera representatives during the analyses (Guseska et al. 2012), respectively alter the ratio between Copepoda and Cladocera in favour of the Cladocera, the especially high quantitative presence of the species *D. cucullata*, typical of eutrophic lakes (Henssen et al. 1995), implies that there are significant changes in the zooplankton community, and hence changes of the water quality in Lake Prespa pelagial.

The recorded results, as well as the chlorophyll *a* analysis (Patceva and Mitic 2010show that negative processes took place in the Lake Prespa in the last several years., that result in the increase of the lake's trophic state. According to Patceva and Mitic (2010), Lake Prespa is in a process of eutrophication and the water level of Lake Prespa has a large effect on its water quality.

The results for the calculated saprobic index for the pelagic part of the Lake are given in the Table

Table 1 Saprobic index in the pelagic zone of Lake Prespa

Species	G. Kl	s
Rotifera		
Filinia longiseta (Ehrenberg, 1834)	II, III	2.3
Kellicottia longispina (Kellicott, 1879)	I, II	1.4
Keratella cochlearis (Gosse, 1851)	II	1.9
Trichocerca capucina (Wierz. et Zach., 1893)	I, II	1.5
S		1.77
Cladocera		
Bosmina longirostris O.F.Müller, 1785	I, II	1.6
Daphnia cuculata G.O. Sars, 1862	I, II	1.7
S		1.65
Copepoda		
Arctodiatomus steindachneri Richard, 1897	I, II	1.5
Mesocyclops leuckarti Claus, 1857	II	1.7
S		1.57
Mollusca		
Dreissena polymorpha (larvae) (Pallas, 1771)	II	1.9
S		1.9

Legend:

s – saprobic index of species

S – saprobic index of locality

G.Kl – water quality

Most of the noted species in the pelagic zone of Lake Prespa were oligo-ß-mesosaprobic and ß-mesosaprobic indicators.

The saprobic index was between 1.57 and 1.9, which corresponds with oligo-ß-mesosaprobic waters. The highest values were recorded for rotifers as more sensitive organisms and good indicators of water quality, as well as for the Dreissena larvae.

6.2. Littoral zone of Lake Prespa

The littoral zone of the lake can be regarded as a very complex area with multiple interactions between zooplankton, biotic and abiotic factors forming a continuous cycle that influences the entire lake ecosystem. Littoral zooplankton communities consist of epiphytic, benthic and planktonic rotifers, copepods and cladocerans.

6.2.1. Rotifera

Inventories of rotifers are important for evaluating the environmental changes and understanding the functional properties of freshwater ecosystems. They are usually considered to be useful indicators of water quality and of the trophic status (Gannon and Stemberger 1978; Sladecek 1983; Marneffe et al. 1995, 1997, 1998; Duggan et al. 2001). Indeed, due to high population turnover rates, rotifers are particularly sensitive to changes in water quality (Sladecek 1983). Their community structure not only allows estimates of the level of pollution, but it can also indicate the trend of the general conditions over time.

The list of Rotifera species recorded in April, 2013, their distribution and saprobic indices, as well as the index of diversity, are presented in Table 2.

Table 2 List of Rotifera species (saprobic indices and Shannon-Wiener index in different localities)

Rotifera species	s	S_i	NW Littoral of Ezerani	Ezerani littoral	NE Littoral of Ezerani	Otesevo
Notholca acuminata (Ehrenberg, 1832)	o-β	1.4	-	+	+	-
Notholca squamula (Müller, 1786)	o-β	1.5	-	-	-	+
Cephalodella forficula (Ehrenberg, 1831)	β	1.8	-	+	-	-
Cephalodella gibba (Ehrenberg, 1830)	β	2.0	+	-	+	+
Trichocerca tenuior (Gosse, 1886)	o-β	1.4	-	+	-	-
Keratella cochlearis (Gosse, 1851)	β-o	1.7	+	+	-	-
Squatinella lamellaris f. *similis* Wulfert, 1960	o-β	1.4	-	+	-	-
Lecane closterocerca (Schmarda, 1859)	β	2.1	-	-	+	-
Trichotria pocillum (Müller, 1776)	β-o	1.6	-	-	+	-
Trichotria tetractis (Ehrenberg, 1830)	β-o	1.6	-	-	+	-
Polyarthra vulgaris Carlin, 1943	β	2.1	-	-	+	-
Encentrum sp. Ehrenberg, 1838	β-o	1.7	-	-	+	-
Euchlanis dilatata Ehrenberg, 1832	β-o	1.9	+	-	-	+
Synchaeta pectinata Ehrenberg, 1832	β-o	1.7	-	-	-	-
Bdelloidea Hudson et Gosse, 1886	β	2.2	+	+	+	+
S			1.95	1.65	1.84	1.9
Shannon-Wiener index (*H'*)			1.38	1.72	2.07	1.38

Legend

s – saprobic degree

Si – individual saprobic index

S – saprobic index of locality

15 Rotifera species were found In the littoral zone of Lake Prespa.. The ecological features of the recorded species show that most of them are cosmopolitan and littoral inhabiting aquatic macro–vegetation.

As it is shown in Table 2, most of the noted species were characteristic for ß–mesosaprobic to oligosaprobic waters and ß–mesosaprobic waters (Sladecek 1983). The calculated Index of saproby (S) showed a ß–mesosaprobic quality of water in all of the analysed localities.

The Shannon-Wiener index (*H'*) that was used, showed the highest diversity values for Ezerani NE.

6.2.2. Crustacea

17 Crustacea species were found in the littoral zone: 12 cladocerans and 5 copepods. The list of species is given in Table 3.

Table 3 List of Cladocera species (saprobic indices and Shannon-Wiener index in different localities)

Registered species	G. Kl	s	Ezerani	Ezerani NW	Ezerani NE	Otesevo
CLADOCERA						
Sida cristalina (O. F. Muler)	I, II	1.4	-	1	1	1
Ceriodaphnia quadrangula (O. F. Muler)	I, II	1.4	-	1	-	1
Simocephalus vetula (O. F. Muler)	II	1.8	1	-	-	-
Bosmina longirostris (O. F. Muler)	I, II	1.6	-	3	-	1
Macrotrix laticornis (Fischer)	II	1.7	3	-	-	-
Acroperus harpe (Baird)	I, II	1.4	1	3	3	1
Alona rectangula Sars	I, II	1.6	-	-	-	1
Alona quadrangulla	I, II	1.6	1	1	1	-
Chidorus sphaericus (O. F. Muler)	I, II	1.8	1	3	5	3
Leydigia acanthocercoides (Fischer)	I, II	1.4	1	-	-	1
Pleuroxus aduncus (Jurine)	I	1.2	1	-	1	1
Rhynchotalona falcata (Sars)	I, II	1.2	1	3	1	1
COPEPODA						
Arctodiaptomus steindachneri (Richard 1897)	I,II	1.5	1	5	5	1
Macrocyclops albidus (Jurine, 1820)	II	2.0	-	1	1	1
Eucyclops macruroides (Lilljeborg, 1901)	I	1.2	1	3	3	1
Megacyclops viridis (Jurine, 1820)	I,II	1.7	3	1	1	1
Mesocyclops leuckarti (Claus, 1857)	II	1.7	1	1	-	1
S			1.56	1.5	1.52	1.54
Shannon-Wiener index (*H'*)			2.36	2.31	2.06	2.56

Legend:

s – saprobic index of species

S – saprobic index of locality

G.Kl – water quality

The most abundant species were *Acroperus harpae, Chidorus sphaericus, Bosmina longirostris, Rinchotalona falcata, Eucyclops macruroides.*

In this period, in the littoral zone of the lake, the domination of cladocerans is an evidence of the intensive process of mineralization. In the oligotrophic waters with low concentrations of bacteria– and phytoplankton as food suitable for filtration, the Cladocera representatives occur with low density, which is not the case in Lake Prespa, especially in this period of the year.

Most of the cladocerans are able to filtrate, 27 millions of bacteria in 24 hours, on average, and have a significant role in the filtration and auto purification of continental waters. Their mortality increases the concentration of the bacteria which is one more reason for the intensive process of eutrophication of the lake.

Some Crustacea species have a high sensitivity to environmental changes and can quickly respond to various types of impacts. This makes them good bioindicators for the evaluation of water quality.

The registered species (Tab. 3) are characteristic for the I,II and II water category (according to Pantle and Buck 1955). The saprobic index was 1.55 which corresponds to oligo–b- mesosaprobic waters.

The employed Shannon-Wiener index (*H'*) showed the highest diversity values for Otesevo.

From the results of the analyses, it can be concluded that the Lake Prespa as one of the oldest and most important lakes in the world, should be protected from the eutrophication process. Additionally, its rich and characteristic living world should be protected as well.

7. Recommendations

The importance of the zooplankton as a significant component in the food chain of the lakes as well as the fact that zooplankton in aquatic ecosystems play an important role in determining the water quality and the stability of the ecosystem, leads to the need of permanent, long–term analyses of this group of organisms.

Rapid changes in the littoral community structure occur over short distances and time intervals, therefore, the understanding of the system requires detailed analyses on both microspatial and short–term temporal scales.

In order to obtain a complete picture of the changes in Lake Prespa's zooplankton community and its ecosystem in general, it is very important to conduct comprehensive, continuous monitoring of all the biological components as an inextricable part of the lake ecosystem.

8. Summary

In the last decade analysis of Lake Prespa's zooplankton imply that there is a significant alteration in zooplankton, not only in terms of its qualitative, but also its quantitative composition, which, in its part, contributes to the structural modifications of the zooplankton components Copepoda, Cladocera, Rotifera.

The density of the total zooplankton in the Lake Prespa pelagic region reached higher values in comparison with the spring period 2008–2009.

During the investigated period, according to the quantity, the Copepoda representatives were dominant, especially calanoid *Arctodiaptomus steindachneri* and the larval nauplial and copepodid stages which are presented with great numerical values.

The cladocerans in this period were presented with lower numerical values. The Cladocera quantity reaches high values during the summer period. According to the analyses in the period between 2008–2009 the numerical values of Cladocera were relatively higher than in the previous analysed

periods.

Most of the noted species in the pelagic zone of Lake Prespa were oligo-ß-mesosaprobic and ß-mesosaprobic indicators. The saprobic index was between 1.57 and 1.9, which corresponds with oligo-ß-mesosaprobic waters.

In the littoral zone of Lake Prespa 15 Rotifera species were found, which is characteristic for ß–mesosaprobic to oligosaprobic waters and ß–mesosaprobic waters and 17 Crustacea species characteristic for the I,II and II water category. The index of saproby (S) showed a ß–mesosaprobic quality of the water in all of the investigated localities.

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REPORT

MICROBIOLOGICAL INVESTIGATIONS OF THE WATER OF LAKE PRESPA AND RIVER GOLEMA

Transboundary Monitoring program

Prepared by Dr. Lence Lokoska
Hydrobiological Institute, Ohrid, R. Macedonia

June, 2014

Contents

List of abbreviations

ALPA - Andrade Lactose Peptone Aqua

APHA – American Public Health Association

CCA - Chromocult Coliform Agar

CFU – Colony Forming Units

MPN - Most Probable Number

TC - Total Coliforms

TFC - Total faecal coliforms

WHO – World Health Organization

WWTP “Ezerani” - Waste Water Treatment Plant “Ezerani”

1. Introduction

The examination of microbiological water quality is obligatory for use-related aspects as such for drinking water, irrigation or recreation. Although microbiological parameters are not standard parameters according to the requirements of the Water Framework Directive (WFD) they are useful to assess the impact especially of pollution causes by domesic wastewater and other organic pollution from diffuse sources. Furthermore analysis of microbiological standard parameters (e.g. fecal indicator bacteria) is obligatory according to the EC-Surface & Drinking Water Directive and the EC-Bathing Water Directive.

Even when biological and chemical water quality is acceptable, microbiological parameters can indicate serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation (Kavka et al. 2002).

The water resources in the Prespa valley are used for different purposes: water supply for populated regions in the valley, water for industrial capacities and irrigation of agricultural regions in all three countries. Beside agriculture use tourism and fishery have economic importance in the lake basin.

Water supplying systems, whose fountains are waters from Macro Prespa Lake, are built in v. Stenje, summer camps Carina and Otesevo, water supplying systems of 18 villages in R. Greece and 12 villages in Albania. Irrigation system for agricultural areas in Macedonia uses water from Macro Prespa Lake. Two pumping stations, one in the village Asamati and the other in the v. Sirhan, were used to water Prespa field. Irrigation system for agricultural areas in R. Greece uses water from Micro Prespa Lake. Irrigation system for agricultural areas in Billis-Corca valley, R. Albania through artificial channel of Canyon Grlo, uses water from Micro Prespa Lake (Chavkalovski, 2000; Sherdenkovski, 2000).

For a long period of time the water level was on peak elevation of 849 m to 850m above the sea level. This period starts from 1951 to 1962 and then from 1970 to 1986. Thanks to the constant water level the coast and surrounding were beautiful with attractive sandy beaches (Sherdenkovski, 2000).

The estimation of bacteriological quality of surface waters based on classical sanitary indicators (total coliforms, faecal coliforms, fecal streptococci) may not reflect their safety for the health of bathing people and/or using water for drinking and household purposes. Numerous human diseases having bath in rivers, lakes, pond and coastal sea waters in the area of river and sewage inflow, swimming, swimming pools are associated with the presence of opportunistic pathogens from *Pseudomonas, Aeromonas, Staphylococcus* and other microorganisms groups, being able to generate infections by contact with skin, mucous membrane, nasopharyngeal cavity, respiratory ducts, eyes, ears and urogenital passages. Pyogenic infection of injuries, meningitis, urinary system, respiratory system, inflammation of the middle ear and eyes are typical diseases caused by contaminated water where Pseudomonas are found (Pellet et al. 1983; Niewolak and Opieka 2000). Coliforms are therefore now being supplanted by more specific indicators. These include *Escherichia coli* and enterococci which are good indicators of gastrointestinal disease, and Pseudomonas which correlates well with ear and skin infections (Warrington and Jonson 2001).

Wound infections, peritonitis, meningitis, endocarditis, septicemia, corneal ulcers, nosocomial infections, urinary tract infections, gastroenteritis of people who bathe and/or use water in other ways are caused by Aeromonas (Kersters et al. 1996, Kuhn et al. 1997). Aeromonas is aquatic, fresh water bacteria that were identified during the last century as responsible of infectious processes in aquatic animals: amphibians, reptiles, fish, snails and others (EPA Office of Water) 2006. Aeromonas numbers range between 1000–10000 bact./100ml normally in waters not affected by sewage pollution. They can occur in numbers up to 100000 bacteria in fresh water affected by sewage pollution. However standards have been established in Europe for numbers of Aeromonas in drinking water:

Water after leaving the treatment plant should have no more than 20 bact/100ml

In the distribution system up to 200 bact./100ml is acceptable.

The purpose of these investigations was to establish the water status of Lake Prespa and its main tributary River Golema according to microbiological aspects (water faecal pollution and water contamination by organic matter) and to assess the impact of the WWTP Ezerani.

To facilitate the interpretation of microbiological water quality data the results were classified according to EC-Bathing water quality directive 2006/7/EEC considering actual expert proposals (Kavka &Poetch, 2008).

2. Parameter and sampling points

Detailed knowledge of faecal pollution in aquatic environments is crucial for watershed management activities in order to maintain safe waters for recreational and economic purposes (Farnleitner et al. 2001). The concentrations of heterotrophic bacteria (heterotrophic plate count, 22^{0}C) correspond commonly with contamination by organic matter (Kohl, 1975).

Parameters of sanitary aspects of the conditions of the water (coliform bacteria, fecal indicators) for determination of its hygiene status: Total Coliforms bacteria (TC), Total Faecal Coliforms (TFC), Enterococci, *Escherichia coli,* Pseudomonas, *Clostridium perfringens,* Aeromonas was investigated.

Furthermore bacteria indicatory of water contamination by organic matter, heterotrophic bacteria was investigated (Colony count 22^{0}C - Aerobic psihrophilic bacteria and Colony count 37^{0}C -Aerobic mesophilic bacteria).

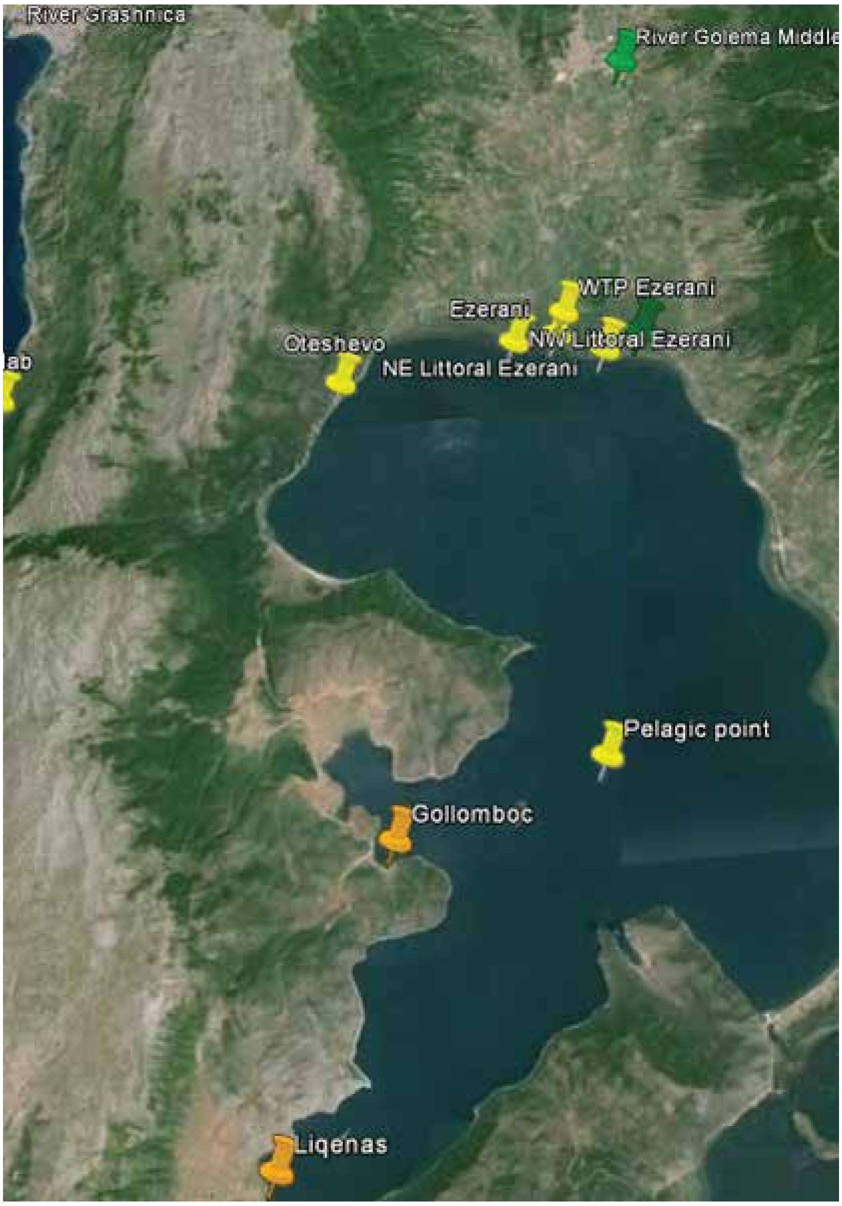

Figure 1. Sampling site Lake Prespa and River Golema

Water samples were collected from following sampling sites on the lake:

MK I WWTP Ezerani (N 41o 00'962; E 21o 01'643);

MK II NW Ezerani (N 41o 00'131; E 20o 56'683), the bottom is very fine sand and mollusc shells, sampling point is near to the reed beds;

MK III NE Ezerani (N 41o 00‘015; E 20o 58‘656), the bottom is very fine sand and sampling point is near to the reed beds;

MK IV Ezerani littoral (N 41o 00‘197; E 20o 57‘741), the bottom is very fine sand and sampling point is near to the reed beds;

MK V Oteshevo (N 40o 58‘692; E 20o 55‘016), the bottom is sand, mud and mollusc shells;

MK VI Pelagic zone (N 40o 57‘949; E 20o 57‘513), the bottom is soft, fine silt

Additionally water samples were collected from the following sampling sites at the River Golema:

Golema 1 River Golema middle course (N 41o 05’040; E 21o 01’324), the bottom is man-made stone bad;

Golema 2 River Golema Inlet (N 41o 00’997; E 21o 01’767), the bottom is mud.

3. Description of investigations

A number of documents contain detailed information on taking samples for analysis of microbiological parameters and their storage and transportation (ISO standard, WHO 2001, 2003, Standard methods -AFA-AWA-WPCF, 2005.

At the water surface, samples for microbiological analyses were collected directly into sterile glass bottles. In deeper water layer, a 2,5 l Ruttner sampler was used.

Samples were collected seasonally from spring 2013 to winter 2014, in four campaigns:

- first campaign in spring
  17.06.2013, Lake Prespa pelagial, littoral and River Golema (middle course and inlet) when the weather was sunny with light southern
- second campaign in summer
  29.07.2013, Lake Prespa pelagial, littoral and River Golema (middle course and inlet) when the weather was sunny
- third campaign in autumn
  30.10.2013, Lake Prespa pelagial, littoral and River Golema (middle course and inlet) when the weather was sunny
- fourth campaign in winter
  26.01.2014, Lake Prespa pelagial, littoral and River Golema (middle course and inlet) when the weather was snow, wind with clouds

4. Transport and storage of samples

The water samples for microbiological analyses were stored in 500 ml sterile glass bottle, and transported to the laboratory in the hand refrigerator at low temperature, and analysed within a maximum of 18-24 h after collection.

5. Analysis

Microbiological indicators of sanitary quality and indicators for organic assessment of contamination were analyzed using standard procedures (Official Gazette of R. Macedonia, number 18/99; Official Gazette of R. Macedonia, number 46/2008); EU-Bathing Water Directive 2006/7/EC and repealing directive 76/160/EEC, EU-Surface & Drinking Water Directive 75/440/EEC, ISO standard, WHO, Standard methods (AFA-AWA-WPCF, 2005).

The following microbiological indicators for Drinking water quality and Bathing water quality ware analyzed: Total Coliforms (TC), Total faecal coliforms (TFC), Enterococci, *Eschericha coli*, Pseudomonas, *Clostridium perfringens*, Aeromonas and heterotrophic bacteria (colony count 22 and 37^{0}C).

Generally *E. coli* and coliforms are used as indicators of contamination of waters, but recent research has shown that Aeromonas are equally important as a measure of water quality because they are implicated in a range of diseases of animals and humans. Pseudomonas correlates with ear and skin infections (APHA-AWA-WPCF, 2005).

The methods for microbiological analyzes are according to Standard Methods (for the examination of water & waste water APHA-21st Edition, 2005) and according to ISO standards, Rodina (1971), Kohl (1975), Kavka (1994)...

Bacteriological analyses

The heterotrophic plate count includes all of the microorganisms that are capable of growing in a nutrient-rich solid agar medium:

- Total count (CFU/1ml or bact/ml) of heterotrophic bacteria (psychrophilic) were determined on after 5-7 days incubation at 22^{0}C, in plate count nutrient agar,
- Total count (CFU/1ml) of heterotrophic bacteria (mesophilic) - in plate count nutrient agar after 72 h incubation at 37^{0}C.

Total coliforms (TC) and *E. coli* were determined according membrane filter method, using Chromocult Coliform agar (CCA, Merck) after incubation 24 h on 37^{0}C. All blue colonies which developed on CCA were accounted as faecal coliforms and *E. coli* and with red are accounted as total coliforms (Farnleitner et al. 2001). This method is approved and accepted as alternative method according to EU water directive and compared to the reference method of the ISO9308-1.

Total faecal coliforms (TFC) were determined according membrane filter method, using Chromocult Coliform agar (CCA, Merck) after incubation 24 h on 44^{0}C, and Multiple-Tube Fermentation procedure (now referred to as the Most Probable Number or MPN procedure on ALPA (Andrade Lactose Peptone Aqua). The most probable number of coliformn bacteria (MPN) was determined by fermentation test on ALPA - liquid medium. The test tubes with observed change of color and presence of gas in the Durham tubes were evidenced as positive (APHA-AWWA-WPCF, 2005).

Enterococci were cultivated on Bile esculin agar (membrane filter technique). Cultural characteristic, after 18-24h at 37^{0}C with brown colonies and luxuriant growth.

For Pseudomonas enumeration 100 ml water was filtered through sterile membrane filter. Each membrane filter was placed on a plate with Pseudomonas (Cetrimide) agar after 72h incubation at 37^{0}C. *Pseudomonas aeroginosa* colonies are fluorescent yellow-green.

Selective Aeromonas medium was used for isolation and enumeration, with membrane filtration and plate method after 24h incubation at 37^{0}C.

Clostridium perfringens was determined on the Wilson-Blair medium in pasteurised (80^{0}C/10min) water samples, after 18h incubation at 37^{0}C. *Cl. Perfringens* produces black colonies.

Classification was performed with class limit values shown in Table 1, proposed by Kavka and Poetsch (2002), and used in the Joint Danube Survey (Kavka et al., 2006).

Table 1: Class limit values for microbial pollutions of rivers assessed by bacteriological standard parameters according to Kohl (1975, modified), Kavka & Poetsch (2002, modified) and in consideration of EU-Bathing water quality directive 2006/7/EEC (target values for E. coli and enterococci concerning a good microbiological bathing water quality = target values in this table) and directive 76/160 EEC (imperative value for total coliforms concerning bathing water quality = target value in this table);

TV=Target Values; FS=faecal streptococci

a) faecal pollution,

Classification of faecal pollution		CLASS				
		I	II (TV)	III	IV	V
Parameter	Faecal pollution	little	moderately	critical	strongly	excessively
Escherichia coli (Fecal coliforms)	In 100 ml water	≤ 100	> 100 -1 000	> 1 000 -10 000	> 10 000 -100 000	>100 000
Intestinal Enterococci (FS)	In 100 ml water	≤ 40	> 40 -400	> 400 -4 000	> 4 000 -40 000	>40 000
Total Coliforms	In 100 ml water	≤ 500	> 500 -10 000	> 10 000 -100 000	> 100 000 -1000 000	>1000 000

pollution by easily degradable organic matter

Classification of organic pollution		CLASS				
		I	II	III	IV	V
Parameter	Organic pollution	little	moderately	critical	strongly	excessively
Heterotrophic Plate Count 22 ^{0}C	In 1 ml water	≤ 500	> 500 -10 000	> 10 000 -100 000	> 100 000 -750 000	>750 000

6. Evaluation

6.1. Total coliform bacteria

The term total coliform bacteria refer to a number of bacteria including *Escherichia, Klebsiella, Citobacter and Enterobacter*. They are associated with feces of warm-blooded animals and are also present in soil. Total coliforms are not specific indicators of faecal pollution (Ashbolt et al, 2001), but can provide basic information on source water quality.

It is evident that the discharge from WWTP Ezerani have much higher coliform count (during all four seasons) than other localities, except River Golema (middle course) where is a maximum of 1 352 000 bact/100ml in the autumn period. Total coliform number in Lake Prespa littoral and pelagial water ranged from 0 to 118 bacteria in 100ml, depending on sampling season and locality.

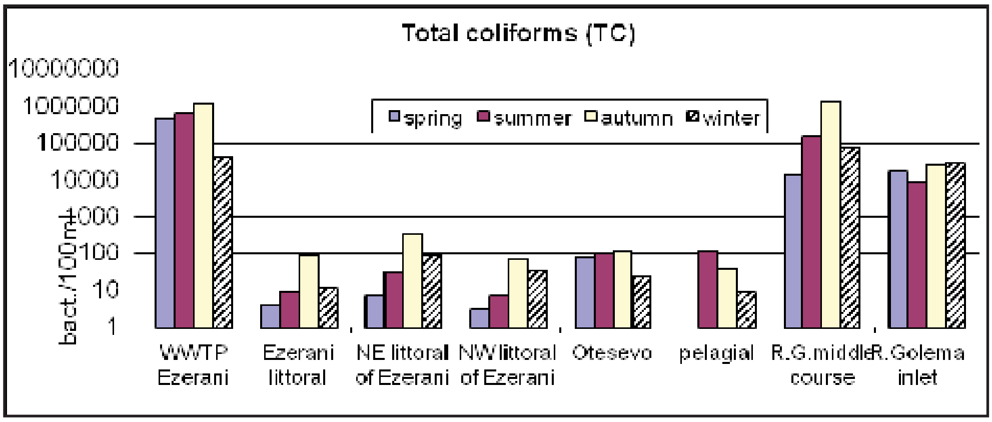

Figure 2: Seasonal fluctuation of total coliforms in the water of Lake Prespa and River Golema

According to presence of the total coliforms, Kavka & Poetsch (2002) classifications, the water of Lake Prespa is mainly unpolluted; at River Golema middle course is moderately (spring, winter-III class), strongly (summer-IV) and excessively polluted (autumn-V class); at River Golema inlet is mainly critical polluted III class); the outflow of the treated water from Treatment Plant "Ezerani" is critical (III) in winter, strongly (IV) in spring and summer and excessively polluted in autumn.

According to presence of the total coliforms and EU-Bathing water quality directive (1976/160/EEC), Lake Prespa is with excellent quality.

6.2. Total fecal coliform bacteria

Fecal coliform bacteria are a subgroup of the total coliform group. They are indicators of fecal contamination because they are restricted to the intestinal tract of warm-blooded animals. Their use enables separation of bacteria from soil and fecal origin.

These bacteria are the most presented in the outflow water of WWTP Ezerani, with maximum of 768 000 bact/100ml (autumn). High values are recorded in the water of the river with a maximum of 617 000 bact./100ml (River Golema middle course- autumn). These bacteria are not registered in the Lake pelagial water and in littoral water, except at NE Ezerani in winter. In the water at Oteshevo these bacteria is not present only in autumn.

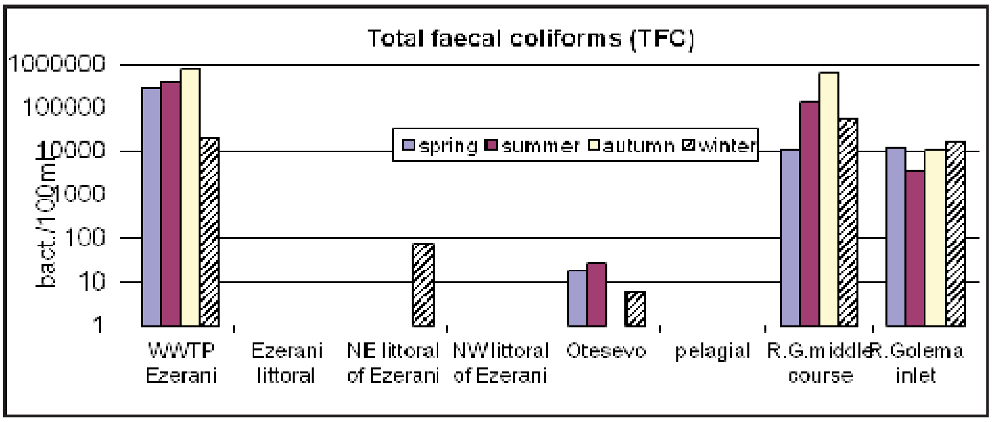

Figure 3: Seasonal fluctuation of total fecal coliforms in the water of Lake Prespa and River Golema

6.3. *Escherichia coli*

Escherichia coli is a type of fecal coliform and is an indicator of fresh fecal pollution. It is evident from the results that relatively high values of *Escherichia coli* are registered in the water of WWTP (632 000bact./100ml) and River Golema (608 000 bact./100ml). In the Lake Prespa E. coli was evidenced only at Oteshevo during spring (3 bact/100m) and summer (5 bact/100m) and at NE littoral of Ezerani just in winter (53 bact/100m).

Escherichia coli occur in high numbers in human and animal faeces, sewage and water subject to recent faecal pollution. Water temperatures and nutrient conditions present in aquatic ecosystems are highly likely to support the growth of these organisms.

It is generally accepted that E. coli and other coliform bacteria can survive in aquatic environments for at least several weeks, depending on the nutriment availability, pH, and water temperature.

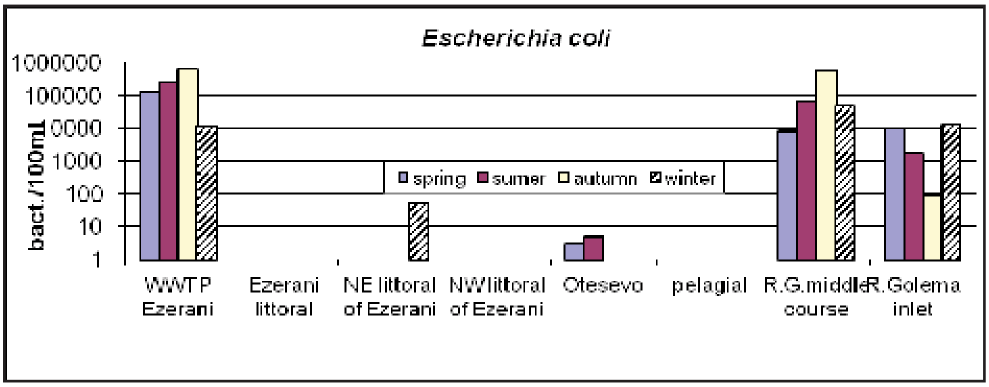

Figure 4: Seasonal fluctuation of Escherichia coli in the water of Lake Prespa and River Golema

For better evaluation of fecal pollution, the number of fecal coliforms *(E.coli)* was also used in investigations. This parameter indicated Ist class water quality in the Lake Prespa, mainly unpolluted; River Golema middle course is critical in spring (III), strongly in summer and winter (IV class) and excessively (V class) in autumn (Kavka & Poetsch 2002, EU-Bathing water quality directive 76/160/EEC, 2006/7/EEC); the outflow of the treated water from Treatment Plant "Ezerani" is mainly excessively polluted (V) and strongly polluted (IVclass in winter).

According to the presence and the numbers of *E. coli* and EU-Bathing water quality directive (1976/160/EEC, 2006/7/EEC), Lake Prespa is with Excellent quality.

6.4. Enterococci

Enterococcus (Fecal streptococci) is another fecal bacteria and is the most sensitive of the indicator bacteria. Important advantages of this group are that they tend to survive longer in water environments than E. coli (or thermotolerant coliforms), are more resistant to drying and are more resistant to chlorination.

Intestinal enterococci are typically excreted in the faeces of humans and other warm-blooded animals. Some members of the group have also been detected in soil in the absence of faecal contamination. Intestinal enterococci are present in large numbers in sewage and water environments polluted by sewage or wastes from humans and animals.

The water of the WWTP Ezerani (21000 to 168 000 bact./100ml) and River Golema (900 to 95000 bact./100ml) contain much more enterococci in comparison with the littoral region of Lake Prespa. In the water of Lake Prespa pelagic region enterococci were not registered, just in autumn.

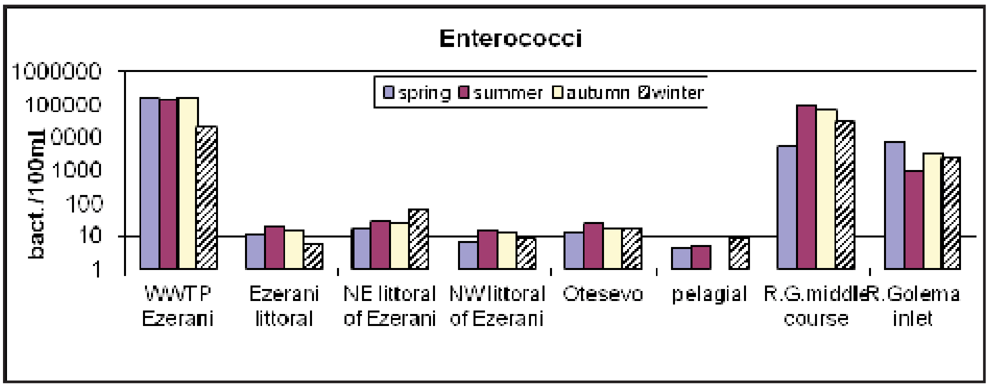

Figure 5: Seasonal fluctuation of Enterococci in the water of Lake Prespa and River Golema

The number of enterococci detected in Lake Prespa shows that pelagial and littoral water is in I class or little contamination to moderately at NE littoral of Ezerani. The water of River Golema middle course is in IV, V class (strongly to excessively contaminated); River Golema inlet is mainly critical polluted (III class) and strongly polluted (IV). The outflow of the treated water from WWTP Ezerani is mainly excessively polluted (V) and strongly polluted (IV class in winter).

According to the presence and the numbers of enterococci, and EU-Bathing water quality directive

(1976/160/EEC, 2006/7/EEC), Lake Prespa is with Excellent quality.

6.5. Pseudomonas

Pseudomonas aeruginosa is a common environmental organism and can be found in faeces, soil, water and sewage. It can multiply in water environments and also on the surface of suitable organic materials in contact with water.

Pseudomonas was identified in the water of all investigated locality of Lake Prespa during the all four seasons. The highest number of this bacteria characterized locality WWTP Ezerani (15000 bact./100ml). Number of Pseudomonas in the water of River Golema ranged from 20 to 1300 bact./100ml, and in the water of Lake Prespa from 3 to 52 bact./100ml (NE of Ezerani).

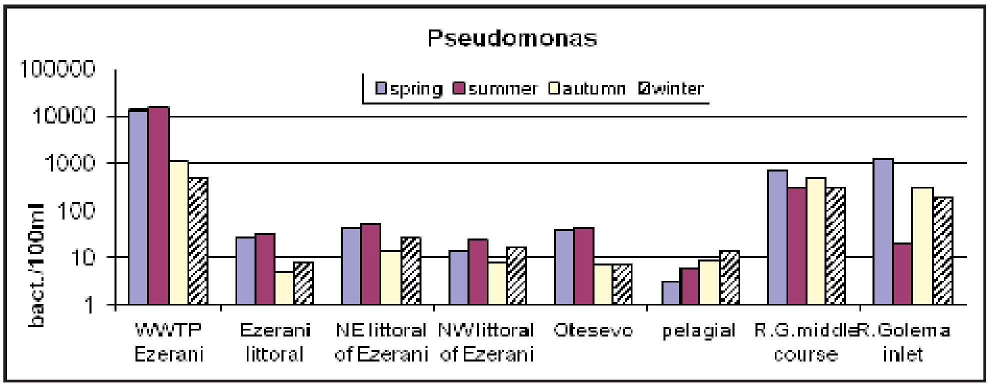

Figure 6: Seasonal fluctuation of Pseudomonas in the water of Lake Prespa and River Golema

6.6. Aeromonas

Generally *E. Coli* and coliforms are used as indicators of contamination of waters. But recent research has shown that Aeromonas are equally important as a measure of water quality because they are implicated in a range of diseases of animals and humans. Aeromonas was registered in all investigated localities during all four seasons. The highest number of bacteria characterized locality River Golema middle course (912 000 bact/100ml –autumn) and WWTP Ezeran (296 000 bact./100ml –summer) and the lowest number was recorded at locality Lake Prespa Pelagial (8 bact./100ml –winter).

Aeromonas range between 1000–10 000 bact./100ml normally in waters not affected by sewage pollution. They can occur in numbers up to 100 000 bacteria in fresh water affected by sewage pollution.

However standards have been established in Europe for numbers of Aeromonas in drinking water: Water after leaving the treatment plant should have no more than 20 bact/100ml. In the distribution system up to 200 bact./100ml is acceptable.

Aeromonas species are increasingly recognized as enteric pathogens; they possess several virulence factors associated with human disease, and represent a serious public health concern. These findings demonstrate the presence of potentially pathogenic and multi-drug- resistant A. hydrophila in the surface waters, thereby indicating a significant risk to public health.

Aeromonas are ubiquitous in the aquatic environment (Holmes et al., 1996). Aeromonas spp. cause disease in poikilothermic animals, and occasionally in mammals. Diseases caused by aeromonads represent a significant source of loss to the aquaculture industry.

Aeromonas species are autochthonous (natural inhabitants) to the aqueous environment where they may be pathogenic for poikilotherms (cold-blooded animals).

Continuous monitoring of surface waters is important to identify potential water-borne pathogens and to reduce the health risk caused by the genus Aeromonas.

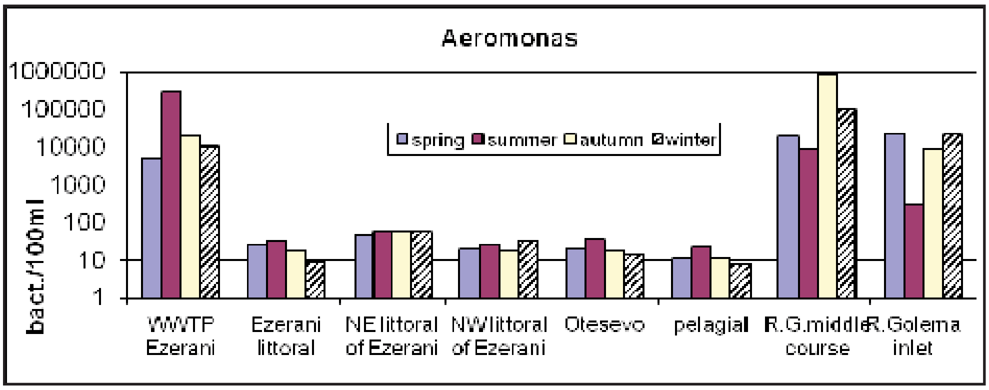

Figure 7: Seasonal fluctuation of Aeromonas in the water of Lake Prespa and River Golema

6.7. *Clostridium perfringens*

Cl. perfringens, is a member of the normal intestinal flora of 13–35% of humans and other warm-blooded animals. Other species are not exclusively of faecal origin. Like *E. coli, Cl. perfringens* does not multiply in most water environments and is a highly specific indicator of faecal pollution.

Cl. Perfringens was not found in the water of Lake Prespa pelagial. In the littoral water was evidenced only in summer period at all localities and in winter at NE of Ezerani. The maximums of this bacteria was registered in the water at WWTP Ezerani (420 000 bact/100ml - summer), and in the River Golema middle course (34 000 bact/100ml - spring).

Clostridium perfringens and its spores are virtually always present in sewage. The organism does not multiply in water environments. *Clostridium perfringensis* present more often and in higher numbers in the faeces of some animals, such as dogs, than in the faeces of humans and less often in the faeces of many other warm-blooded animals. The numbers excreted in faeces are normally substantially lower than those of *E. coli*.

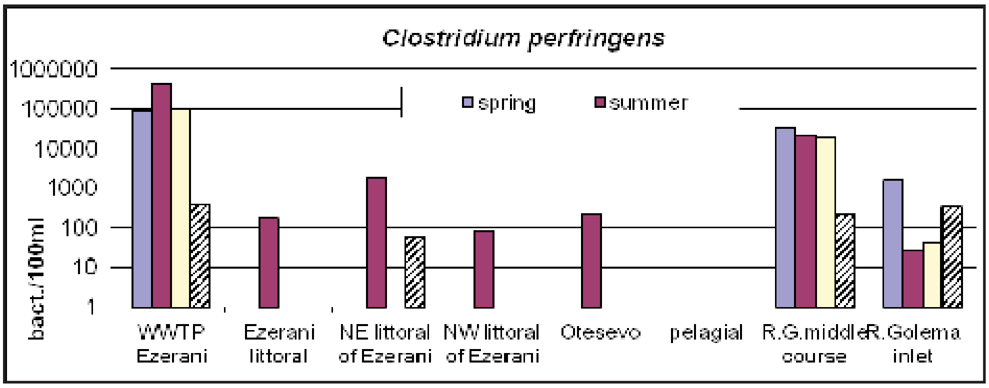

Figure 8: Seasonal fluctuation of Cl. perfringens in the water of Lake Prespa and River Golema

6.8. Heterotrophic bacteria (colony count 22^{0}C and 37^{0}C)

The highest number of heterotrophic bacteria was found in the water of WWTP Ezerani (948 800), and in the River Golema middle course (670 400 bact/ml).

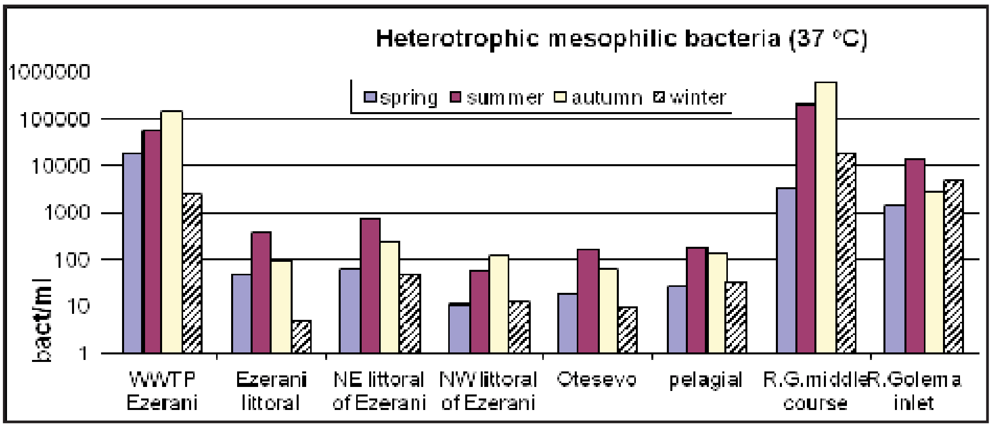

Figure 9: Seasonal fluctuation of Heterotrophic bacteria (370C) in Lake Prespa and River Golema

The number of mesophilic bacteria ranged from 5 bact/ml (at Ezerani littoral -winter) to 752 bact/ml (at NE of Ezerani -summer) in the lake water. The maximal number of this bacteria in the water of River Golema middle course is in autumn (614 400 bact/ml), and in the water of River Golema inlet is in summer (13 700 bact/ml). The number of this bacteria varied from 2410 to 142 400 per ml in the outflow of the treated water from the WWTP Ezerani (winter, autumn). Such a high presence of mesophilic heterotrophic bacteria indicates the possibility of eutrophication, above all eutrophication of anthropogenic origin. This situation is proved with the presence of coliform bacteria and fecal indicator bacteria. The high presence of mesophilic bacteria is in correlation with high presence of coliform bacteria.

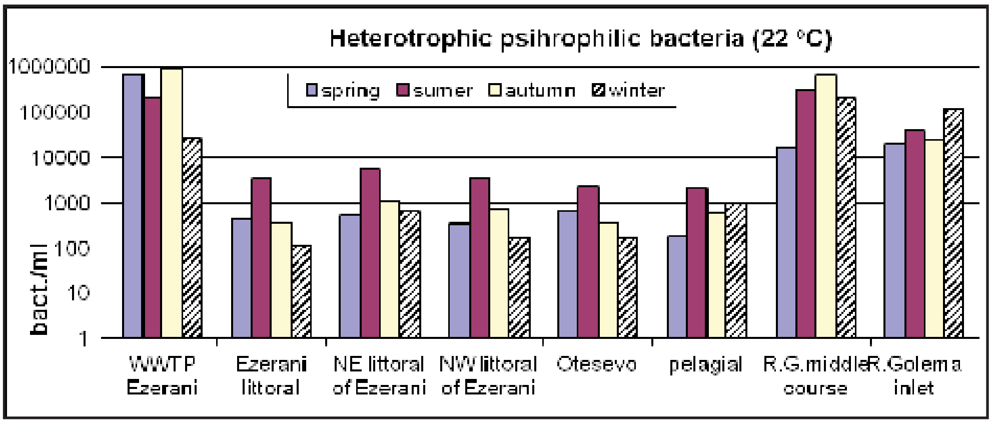

Figure 10: Seasonal fluctuation of Heterotrophic bacteria (220C) in Lake Prespa and River Golema

The number of psychrophilic bacteria varied from 114 bact/ml (at Ezerani littoral -winter) to 5 520 bact/ml (at NE littoral of Ezerani-summer) in the lake water, and from 17 280 bact/ml (spring) to 670 400 bact/ml (autumn) in the water of River Golema middle course.

A smaller number of mesophilic bacteria as of psychrophilic heterotrophic bacteria have been observed.

Generally the number of heterotrophic bacteria increases with the trophy and the organic pollution of water bodies.

The water quality according to the psychrophilic heterotrophic bacteria Lake Prespa (pelagial and littoral) is in I and II class. River Golema middle course is mainly strongly (IV class) and critical (III-spring). River Golema inlet is mainly critical (III class) and strongly (IV winter) (Kohl, 1975).

Potential sources of bacterial contamination include surface water runoff, animal waste, and leachate from sewage disposal systems and improperly disposed of human waste from visitors to the lake and rivers.

7. Classification

Bacteria concentrations are higher in Golema river as in the littoral and pelagial zone of the lake. The discharge from the WWTP Ezerani as well as the run off of Golema river are typical sewerage water with high values of all investigated bacteria.

The interpretation of microbiological water quality data, and the results are classified according Kohl (1975), EU-Bathing water quality directive (1976/160/EEC and 2006/7/EEC) and EU expert proposals -Kavka & Poetsch (2006).

Microbiological determinants indicated a good bacteriological water quality at the sampling sites of the lake. In this localities colony counts of heterotrophic bacteria indicated little (class I) to moderate pollution (class II) by organic matter, and low contamination with fecal bacteria (class I); only enterococci were a little increased.

Results of the microbiological examinations of the discharge of the WWTP Ezerani show remarkable fluctuations of the qualitative and quantitative composition of investigated groups of bacteria. Regarding the numbers of heterotrophic bacteria and total coliforms, outflow water of WWTP is strongly polluted (class IV) to excessively (class V) polluted, except in the winter when is critical polluted (class III). Fecal indicator bacteria (*E. coli*, enterococci) reflect strongly to excessively faecal contamination of water (class IV and V).

The water quality of River Golema is decreasing downstream. In the samples taken in the town of Resen waste communal and industrial water was present in the river. This situation is because the river water (after Resen) is used uncontrolled for irrigation, from May to October.

River Golema middle course, according to total coliforms is critical polluted (class III) in winter and spring, and strongly to excessively polluted in summer and fall (class IV and V). The presence of *E. coli* and enterococci indicated strongly to excessively fecal pollution (class IV and V). Presence of heterotrophic bacteria indicated strongly (class IV) pollution by organic matter.

River Golema inlet - fecal indicator bacteria (*E. coli*) reflects mainly critical (class III –spring, summer, autumn) to strongly fecal contamination of water (class IV –winter). Critical concentrations of enterococci were observed (class III) in all season except in spring when the water is strongly polluted (class IV). Heterotrophic bacteria indicated critical (class III –summer, autumn, winter) to strongly (class IV – spring) pollution by organic matter.

The results of microbiological investigation showed that the water of River Golema –middle course is most polluted.

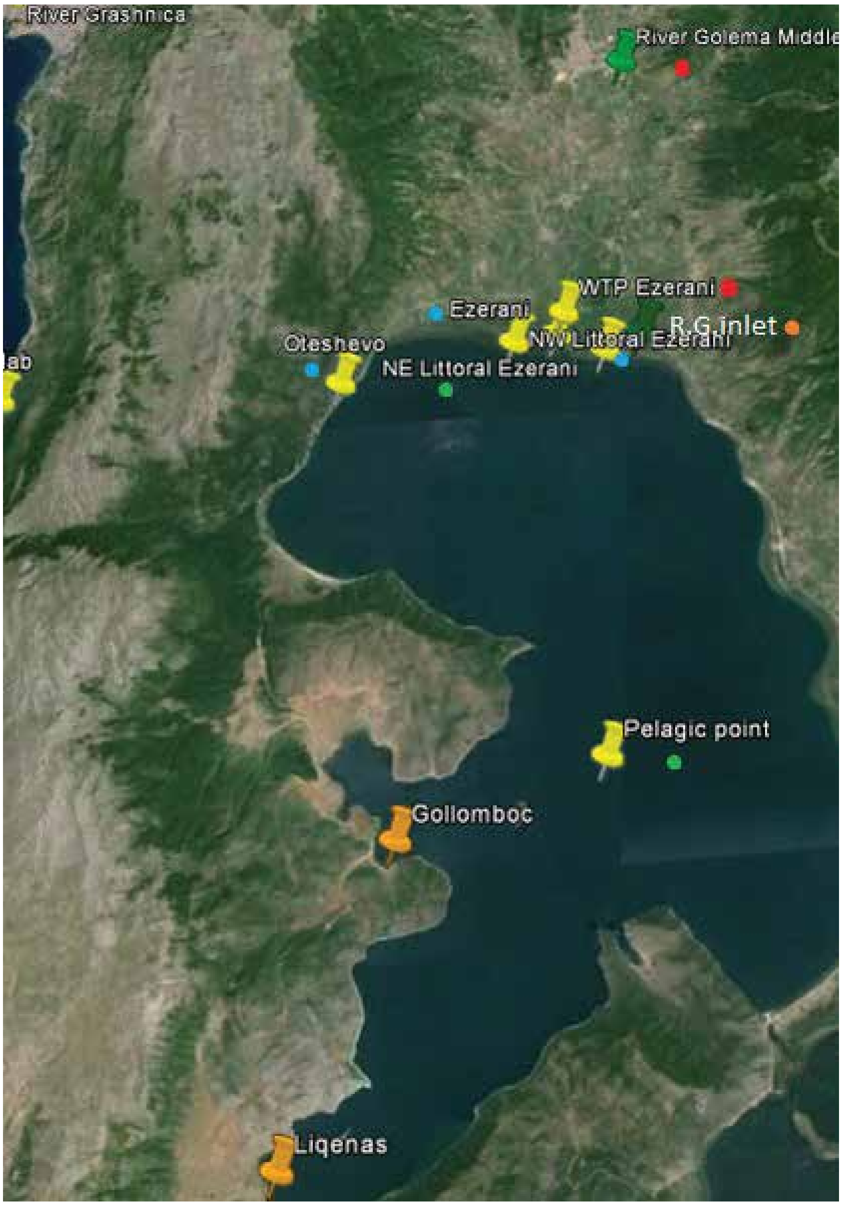

Figure 11: Microbiological map of fecal pollution assessed by the concentrations of fecal coliforms and additionally concentrations of enterococci: blue coloured symbols: class I-little fecal polluted; green; class II-moderately; yellow: class III-critica,; orange: class IV-strongly; red: class V –excessively fecal polluted

During the all sampling campaigns, the highest abundance of Pseudomonas was recorded in the WWTP Ezerani (15 000 bact./100ml) and in the water of River Golema inlet (1 300 bact./100ml) during summer and spring period.

Generally *E. coli* and coliforms are used as indicators of contamination of waters. But recent research has shown that Aeromonas are equally important as a measure of water quality because they are implicated in a range of diseases of animals and humans.

The lowest number of Aeromonas (8 bact./100ml) was evidenced in the water of Lake Prespa pelagial during the winter. Its maximal representation was in the water of River Golema (912 000 bact/100ml in autumn) and in the water of WWTP Ezerani (296 000 bact./100ml) during the summer period, show that this waters are affected by sewage pollution.

The results of the investigations show that the water of Lake Prespa is clear, and can be used for drinking with treatment.

Lake Prespa has Excellent Bathing Water Quality.

The obtained values for fecal coliforms, E. coli, enterococci and Pseudomonas in the water of River Golema, exceed the prescribed values for water quality for irrigation.

8. Recommendations

Continuous and complex investigations of the littoral and pelagial region are necessary, especially the part at Ezerani (littoral zone in front of the wetland Ezerani –before WWTP Ezerani), and lake tributaries to observe the pollution from the WWTP Ezerani.

Microbiological analyses must be included in all investigations performed on Lake Prespa and other aquatic ecosystems, as a good indicator for water quality, water categorization and determination the source of pollution.

Even when biological and chemical water quality is acceptable, microbiological parameters can indicate serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation (Kavka et al. 2006).

Recommendations for Future Microbiological Monitoring:

- Microbiological water quality was described by one proposed assessment method and supported the detection of anthropogenic impacts;
- Microbiological results support ecological and physico-chemical assessment of the natural state and water quality of the Lake Prespa and its tributaries;
- Microorganisms are very sensitive indicators for the detection of fecal and organic pollution caused by raw sewage, municipal waste water treatment plants and diffuse impact from farmland; quantitative results from indicator bacteria facilitate the detection of hot spots;
- Microbiological investigations are obligatory for testing compliance with the requirements for the utilizations of water for drinking, bathing, irrigation and another economic purpose.

9. Summary

Bacteria are ideal sensors for the indication of microbial pollution of surface water ecosystems because of their fast response to changing environmental conditions. Even when biological and chemical water quality is acceptable, microbiological parameters can indicate serious anthropogenic impact characterized by high bacterial counts. The examination of microbiological water quality is obligatory for use-related aspects, such as for drinking water production, irrigation or recreation.

During all four sampling campaigns on Lake Prespa and River Golema, the obtained results for investigated bacteria showed that generally, their values in the river and some part of littoral are with a great variability.

The obtained results for all investigated bacteria showed that generally, their values in River Golema show great variability, and are considerably higher than in the water of Lake Prespa.

Microbiological determinants indicated a good bacteriological water quality at the sampling sites on the lake.

The water quality of Golema River is decreasing downstream caused by the impact of communal and industrial waste water which is not discharged into the sewer. The situation is strengthened by the extraction of river water (after Resen) for irrigation, from May to October.

Lake Prespa has excellent Bathing Water Quality.

The obtained values for faecal coliforms, E. coli, enterococci and Pseudomonas in the water of River Golema, exceed the prescribed values for water quality for irrigation.

Continuous and complex investigations of the littoral and pelagial region are necessary, especially the part at Ezerani (littoral zone in front of the wetland Ezerani –before WWTP Ezerani), and lake tributaries to observe the pollution from the WWTP Ezerani. Furthermore sediment investigations in the river bed and sediments could provide information about the pollution potential.

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