

## MICROBIOLOGICAL ASSESSMENT OF THE LAKES BISTARAC AND VIDARA WATER QUALITY

### Mikrobiološke karakteristike vode jezera Bistarac i jezera Vidara

Snježana Hodžić<sup>1</sup>, Suad Širanović<sup>1</sup>, Aldijana Avdić<sup>1</sup>, Darja Husejnagić<sup>1</sup>, Almedina  
Djedović<sup>1</sup>, Nahida Vuković<sup>1</sup>

#### Abstract

In the spring and summer season of 2014, the change in the number of coliform and heterotrophic bacteria in specific localities of lake Bistarac and lake Vidara were observed and analyzed. Lake Bistarac was formed after the cessation of surface mining in the open pit, while lake Vidara is the artificial lake in the area of Gradacac made in order to protect the industrial zone of flooding. Both of these lakes, placed in Tuzla Canton, are very attractive for tourist and frequently are used for swimming, fishing and recreation. Results of this study showed an increased number of coliform bacteria in both periods, and the presence of *Escherichia coli* as a direct indicator of fecal pollution, while, as an additional indicator of fecal pollution, *Citrobacter* and *Enterobacter* were identified. From the total number of analyzed water samples of the lake Bistarac, 60% of samples belonged into to the second class, and 40% into the first class of water quality. Lake Vidara showed poor quality as the 80% of the analysed samples belonged to the second class and only 20% into the first class of water quality. For the microbiological assessment of the water quality standard microbiological procedures and methods were used. One of the main reasons for poor hygienic state of these two lakes is unregulated sewage network of the villages, restaurants and camping areas placed near the lakes. The increased number of fecal bacteria was found in the water of both lakes, which can be highly dangerous for the human population. Since both lakes are used for recreational purposes it is necessary to establish an appropriate monitoring system on the basis of which is possible to take measures and to protect and improve water quality.

*Key words: water quality, coliform bacteria, fecal pollution, hygienic assessment*

#### INTRODUCTION - Uvod

Repletion with sufficient quantities of good quality water is crucial for the health of the entire population. Water can be natural residence for various microorganisms, while on the other hand the microorganisms can enter the water from the different environments: air, earth, with plants and excreta from human and animal origin. The most significant contaminants of the surface and drinking waters, from the

---

<sup>1</sup> University of Tuzla, Faculty of Science, Department of Biology

aspect of the human health, are the microorganisms originated from human and animal excreta, which contaminate water mainly due to the unsanitary disposal of waste water. (KARAKAŠEVIĆ, 1989). This usually happens during natural disasters and floods. Many pathogens can be found in the water as potential contaminants. The secure indicators of drinking water fecal pollution include: *Escherichia coli*, *Streptococcus (Enterococcus) faecalis*, representatives of *Proteus* genus, while additional indicators are coliform bacteria (*Citrobacter*, *Klebsiella*, *Enterobacter* and *Serratia*), *Clostridium perfringens* and bacteriophages. Some of these pathogens may cause very serious diseases: cholera, typhoid, paratyphoid fever, bacterial dysentery, hepatitis A, poliomyelitis ect (HUKIĆ,1995).

Vidara is an artificial lake in the area of Gradačac (Bosnia and Herzegovina), made in 1971, in order to protect the industrial zone of flooding. This lake is located in the suburb of Gradačac about 2km from the city centre. In a completely natural environment, this lake is a unique example of untapped tourism potential. It is supplied with water from mountain streams and a few small wellsprings of drinking water. Many small forest streams that swell during rainy days also flow into the lake, carrying a large amount of water (web 1).

Lake Bistarac was formed after the cessation of surface mining of lignite in the open pit, located near the town of Lukavac. It is characterized by his advanced stage of progradation and great heterogeneity of its habitats. The lake is supplied by surface water that flows from the northwest side of the lake and has an overflow system, so the water level during the year is relatively constant. Local truism has been developed in this area, so the lake Bistarac is generally known for its modern swimming pools, camping sites, sports grounds and landscaped picnic areas (web 2). The aim of this study was to establish the difference in water quality in the spring and summer season, between pit lake Bistarac and the lake Vidara, since they are both artificial lakes which are currently used for tourism purposes.

#### **MATERIAL AND RESEARCH METHODS – *Materijal i metode istraživanja***

The microbiological quality of water of lake Vidara and Bistarac was analyzed in the spring and summer of 2014. Before each sampling, water temperature has been measured. Samples were taken from five sites (Picture 1,2) on both lakes (east, west, north, south and center of the lake) in the appropriate sterile container volume of 1000 ml and then are transported to the cooling systems to labs, where they have been analyzed. All analysis were conducted in the laboratory for microbiology at the Faculty of Science, University of Tuzla.

Microbiological water analysis included determining the total number of aerobic mesophilic and psychophilic bacteria in 1 ml of water, the most probable number of coliform bacteria in 1000 ml of water, and the determination of coliform bacteria of faecal origin, were carried out according to microbiological procedures determined with the ISO standards (ISO 9308-2:2012; ISO 6222:1999). The "IMVIC" test was used for the determination of coliform bacteria.



Figure 1-2. Bistarac Lake and Lake Vidara (Google Maps) with marked locations of sampling (1-east, 2-west, north-3, 4-south; 5-middle of the lake).

Slika 1-2. Jezero Bistarac i jezero Vidara (Google Maps) sa označenim lokalitetima uzorkovanja (1-istok; 2-zapad; 3-sjever; 4-jug; 5-sredina jezera)

### RESULTS- Rezultati

From water samples taken the lake Bistarac during the spring season, the number of bacterial colonies grown at temperature of 37<sup>0</sup>C was ranged from 85 to 290, and the number of colonies grown at temperature of 22<sup>0</sup>C was ranged from 20 to 220. In the summer season, colony counts were ranged from 80 to 300 at 37<sup>0</sup>C and 115-190 at 22<sup>0</sup>C. The water temperature at the sampling sites and the temperature during the season is shown in Table 1.

**Table1.** The water temperature of the lake Bistarac and the ratio of mesophilic and psychrophilic microorganisms in the spring and summer season

**Tabela 1.** Temperatura vode jezera Bistarac i odnos mezofilnih i psihofilnih mikroorganizama u proljetnoj i ljetnoj sezoni

Locations of the Lake Bistarac	Seasons	Number of colonies at 37 °C	Number of colonies at 22° C	The ratio of mesophilic and psychrophilic microorganisms	Water temperature (°C)
East	Spring	115	20	57,5 : 10	21
	Summer	300	190	15,8 : 10	24
West	Spring	180	60	30 : 10	20,5
	Summer	150	115	13 : 10	23
North	Spring	170	190	8,9 : 10	20
	Summer	130	115	11,3 : 10	23
South	Spring	290	220	13,2 : 10	21
	Summer	80	140	5,7 : 10	23
Middle	Spring	85	100	8,5 : 10	19,5
	Summer	160	160	10 : 10	24

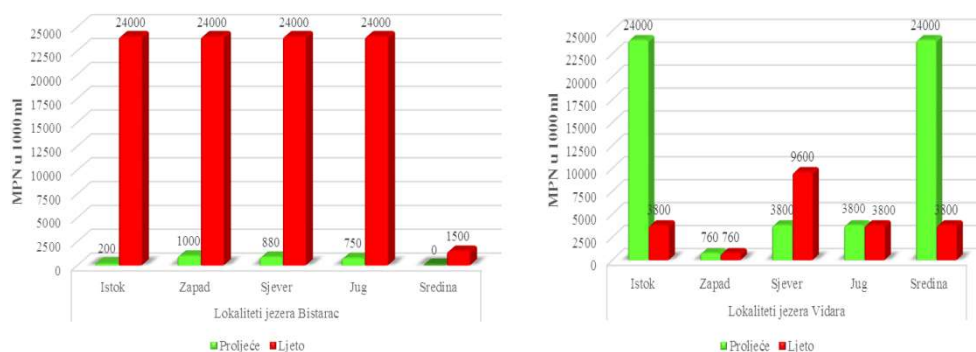
Results obtained by analyzing water samples from the lake Vidara during the spring season, have shown high number of colonies grown at temperature of 37<sup>0</sup> C and 22<sup>0</sup>C (Table 2). Higher number of bacteria was recorded during the summer season where the number of colonies at 37<sup>0</sup>C ranged from 173 to 450 and at a temperature of 22<sup>0</sup>C from 224 to 568. The water temperature and season of sampling is shown in Table 2.

**Table2.** The water temperature of the lake Vidara and the ratio of mesophilic and psychrophilic microorganisms in the spring and summer season

**Tabela 2.** Temperatura vode jezera Vidara i odnos mezofilnih i psihrofilnih mikroorganizama u proljetnoj i ljetnoj sezoni

Locations of the Lake Bistarac	Seasons	Number of colonies at 37 °C	Number of colonies at 22° C	The ratio of mesophilic and psychrophilic microorganisms	Water temperature (°C)
East	Spring	62	106	5,8 : 10	21
	Summer	173	224	7,7 : 10	24
West	Spring	300	300	10 : 10	20
	Summer	249	253	9,8 : 10	24
North	Spring	146	300	4,9 : 10	20
	Summer	178	283	6,3 : 10	24
South	Spring	183	335	5,5 : 10	20
	Summer	450	448	10 : 10	23
Middle	Spring	128	355	3,6 : 10	20
	Summer	262	568	4,6 : 10	24

Maximum value of MPN for the lake Bistarac in the spring season was 1000, while in the summer season much higher values were registered (max MPN was 24 000) (Graph 1). The minimum MPN value for the lake Vidara, was recorded on the west side (760) and the highest on the east side of the lake (24 000). In the summer of MPN values were lower and ranged from 760 (west side of the lake) to 9600 (the north side of the lake) (Graph 2).



**Graph 1 and 2.** MPN values in 1000 ml of water in the spring and summer seasons determined at different sites at the lakes Bistarac and Vidara

**Grafik 1 i 2.** MPN vrijednosti u 1000 ml vode u proljetnoj i ljetnoj sezoni na različitim lokalitetima jezera Bistarac i jezera Vidara

Biochemical characteristics of the summer isolates coliforms from the east, west, north and south sides of the lake Bistarac, indicated the presence of *E. coli*, while in the sample of water from the middle of the lake indicate the presence of *Enterobacter* sp. In the spring season at the sites of the east, west and north isolated *Citrobacter* sp., and at the site south of *E.coli*. From the middle of the lake Bistarac coliform bacteria were not isolated.

Coliform bacteria are identified in the samples taken from the lake Vidara in the spring and summer season. Isolated bacteria are identified as *E.coli* and bacteria of the genus *Citrobacter* and *Enterobacter*.

### DISCUSSION - *Diskusija*

Although the determination of coliform bacteria has much more importance, determination of total number of aerobic mesophilic bacteria in 1 ml water sample, gives an outline picture in assessment of bacteriological quality of drinking water. This data can be of great significance in certain circumstances that may endanger the aquifer such as sudden weather changes (drought or rainfall), field changes, etc. This is very important to emphasize because the microbiological analysis of the lake was carried out in the period after the floods that hit Bosnia and Herzegovina in May 2014. The results obtained by this analysis point to an increased number of aerobic bacteria in all localities on both lakes. The highest number of bacteria (CFU=300), was determined in the samples taken from the east location of the lake Bistarac. This indicates a significantly increased number of bacteria in the water, compared to the results obtained in previous studies (MAŠALA; 2009) (max CFU=145). This increase in bacterial number may have a connection with natural disasters that hit this area a few days before these analyses.

The presence of coliform bacteria of fecal origin was found on all locations of the lake Bistarac, except the central part in the spring season. The most probable number of coliform bacteria has a maximum value in the summer season, where for the east, west, north and south location the MPN was 24 000. This was a significant increase in regard to spring season. However, is part is a bit isolated from the rest of the lake, and the layers of mud and waste that has caused flooding stopped just in this area and significantly contributed to such microbiological picture. The MPN values for the summer season are notably increased due to higher water temperatures, but mostly it can be connected with the beginning of the swimming season on the lake, since one part of the lake rebuilt as a distinctive tourist complex in the summer and is visited by a large number of swimmers. It should be noted that close by the lake, on its south side, there is a restaurant and several houses, whose sewage network is questionable. The information on sewage network regulation is not available. The fact that the presence of these objects affects the microbiological state of the lake, confirms the MPN result, which is quite high in samples taken from the south location the lake (especially during the summer season when the visits to the restaurant are much frequent). The maximum value of the most probable number of coliform bacteria (24000) in our study was significantly different from the recorded value (3800) in the previous studies (MAŠALA, 2009). These results points to the conclusion that there is constant presence of the pollution at this lake, over the last few years.

From the identified coliform bacteria, *E.coli* had a major presence in analyzed samples. These bacteria are considered one of the main indicators of fecal pollution, and their presence in water samples indicates the presence of potentially pathogenic microorganisms (DURAKOVIĆ, 1991). Along with *E.coli*, in water samples taken from various locations on the lake Bistarac, other coliform bacteria were identified: *Citrobacter* sp. and *Enterobacter* sp. This further confirms the fact of recent fecal contamination of the lake at the time of sampling. The major fecal contaminants are the villages located near the lake, with a stable mode of animal husbandry. Soils of these villages are rich in a variety of bacteria of fecal origin, and rinsing these soils by rain or floods, leads to greater pollution of lakes.

Total number of aerobic mesophilic bacteria in water samples from the lake Vidara, showed a significant increase during the summer season, with maximum CFU value of 450 (at the south location). Whereas the number of psychrophilic bacteria was higher than mesophilic (max CFU= 568). The ratio between the number of mesophilic and psychrophilic bacteria is very significant form the aspect of determination of the pollution degree. As the ratio is larger, degree of contamination is higher, which in this case is expressed.

In addition to these data, indicating the contamination of the lake, MPN values riched 24 000 in the spring season. The floods that hit this area in May 2014, largely affected its microbiological picture. Large amounts of rainfall have caused spills of sewage disposal from the local facilities into the lake, as the largest number of such facilities doesn't have regulated hygienic and sanitary infrastructure. Near the lake, also noumerous farmlands and orchards are located, wich are threated with manure.

Manure is rinsing from the soil during the heavy rainfalls and it is draining into the lake.

The fecal contamination of the lake also indicates the findings and identification of coliform bacteria in water samples. In most of the samples, taken from the lake in the spring season, *E.coli* was most commonly presented, along with *Enterobacter* i *Citrobacter* species.

In the summer season this lake is visited by great number of local swimmers, although there is neither urbanized beach nor any significant tourist facilities. Because the lake is located in completely natural surroundings, a large number of campers are attracted and are settled on the shores of the lake during the summer season. Since there is no organized camping resort, disposal of garbage and feces from these camping areas, significantly affects the contamination of the lake Vidara.

Since, the analyses of water quality of the lake Vidara have not been conducted so far, these results can be considered as preliminary.

Based on the MPN value for the coliform bacteria in 1000ml of water, 80% (4/5) of water samples taken from the lake Bistarac, during the spring season, belonged to the class I and 20% (1/5) II class quality. Water quality of the lake Vidara in the spring season was much poorer than the water quality of lake Bistarac, 80% (05/04) of samples belonged to class II and only 20% (1/5) to the class I. Poor water quality of the lake Vidara in the spring season can be attributed to the basic function of the lake which is to protect the industrial zone of flooding. Given that the amount of rainfall in the spring was much higher, is clear why the water quality of lake Vidara was inferior to quality water of the lake Bistarac. During the summer season, water quality of both lakes was even worse; 80% of the water samples from the lake Vidara and all samples from the lake Bistarac, belong to the class II. Poorer water quality of the lake Bistarac compared to the lake Vidara during the summer season can be correlated with the massive use Bistarac lake for recreational purposes.

According to the Regulation of Water Classification (Uredba o klasifikaciji voda), water of the class II can be used for swimming, recreation and water sports, for cultivating less edible fish (cyprinids), and with the usual methods of treatment (coagulation, filtration and disinfection) may be used for beverage and food industry.

### **CONCLUSION - Zaključak**

The lake Vidara, with main function to protect the industrial zone of flooding, showed better water quality during the summer season than in the spring season. On the other hand, the lake Bistarac showed opposite results. Natural disasters (floods), increased human impact, especially during the summer when the swimming season starts, and unregulated hygienic and sanitary infrastructure of the local villages, these were all important factors that affected water quality. Since both lakes are used for recreational purposes there is a need to reduce an anthropogenic influence to a minimum and to establish an appropriate monitoring system on the basis of which it is possible to take measures to protect and improve water quality.

## **REFERENCES- *Literatura***

- DURAKOVIĆ, S. (1991). Prehrambena mikrobiologija, Medicinska naklada, Zagreb.
- HUKIĆ, M. (1995): Bakteriologija dijagnostika, Tuzla: Prvo izdanje.
- INTERNATIONAL ORGANIZATION FOR STANDARDIZATION (2012) Water Quality – Enumeration of Escherichia coli and coliform bacteria- Part 2: Most probable number method. ISO 9308-2:2012. Geneva, ISO.
- INTERNATIONAL ORGANIZATION FOR STANDARDIZATION (1999) Water Quality – Enumeration of Culturable Microorganisms – Colony Count by Inoculation in a Nutrient Agar Culture Medium. ISO 6222:1999. Geneva, ISO
- KARAKAŠEVIĆ, B. (1989): Mikrobiologija i parazitologija, Beograd-Zagreb; Medicinska knjiga.
- MAŠALA, A. (2009): Bacteriological characteristics of the lake waters in the Tuzla canton. Veterinaria 58(3-4) 219-228, Sarajevo 2009
- UREDBA O KLASIFIKACIJI VODA I VODA OBALNOG MORA, „Službeni list BiH“ broj 18/80.
- UREDBA O KLASIFIKACIJI VODA I VODA OBALNOG MORA JUGOSLAVIJE U GRANICAMA SR BIH „Službeni list SR BiH“ broj 19/80 (preuzeta uredba).
- WEB1: [http://www.fmoit.gov.ba/userfiles/file/SUO\\_Sanacije%20deponije%20Visnjik,%20Gradacac\\_2.pdf](http://www.fmoit.gov.ba/userfiles/file/SUO_Sanacije%20deponije%20Visnjik,%20Gradacac_2.pdf). (pregledano: 29.01.2017.)
- WEB2: <http://lukavac.ba/wp-content/uploads/2012/02/Izvjestaj-o-stanju-okolisa-Lukavac.pdf>. (pregledano: 29.01.2017.)



## SAŽETAK

Mikrobiološka analiza vode jezera Bistarac (općina Lukavac) i jezera Vidara (općina Gradačac) vršena je u proljetnom i ljetnom periodu 2014.godine. Uzorci za analizu uzeti su sa pet lokaliteta na oba jezera, a analiza je uključivala slijedeće parametre: određivanje ukupnog broja aerobnih mezofilnih bakterija u 1 ml uzorka vode, i određivanje prisustva koliformnih bakterija (*E. coli*, *Klebsiella*, *Enterobacter* i *Citrobacter*). Rezultati istraživanja pokazali su povećan ukupan broj bakterija kao i veći broj koliformnih bakterija u oba perioda, te prisustvo bakterije *E. coli* kao osnovnog indikatora fekalnog zagađenja, dok su od dodatnih indikatora fekalnog zagađenja koliformnih bacila prisutni: *Citrobacter*, *Enterobacter*. Jedan od glavnih razloga lošije higijenske slike ova dva jezera su velike poplave koje su se desile nekoliko dana prije provedenog istraživanja, ali i mnogi drugi faktori koji doprinose ovakvim rezultatima kao što su sezona kupanja na ovim jezerima, neregulisana kanalizaciona mreža naselja i objekata u blizini jezera, kamp naselja i dr. Prema dostupnim podacima o mikrobiološkim analizama rađenim na jezeru Bistarac ranijih godina, ukupan broj bakterija kao i broj koliformnih bakterija fekalnog porijekla drastično se povećao, što ukazuje na činjenicu da zagađenja na ovom jezeru uzima sve više maha. Mikrobiološke analize na jezeru Vidara nisu prije rađene, tako da dobijene rezultate našeg istraživanja možemo smatrati preliminarnim.

***Corresponding author: Snježana Hodžić, University of Tuzla, Faculty of Science, Department of Biology, Dr. Tihomila Markovića 1, Tuzla 75000, Bosnia&Herzegovina; e-mail: snjezana.hodzic@untz.ba***