

## **Microbiological characteristics and hygienic-sanitary aspect of the water from Black River in the Pelagonija region-North Macedonia**

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### **Abstract**

The presence of microorganisms in the aquatic environment has a variable character and differs from one aquatic ecosystem to another. The main goal of this research was to determine the microbiological characteristics of the water in Black River in the Pelagonija region near the city of Bitola, North Macedonia and thus to determine the quality of water from a hygienic-sanitary aspect. The material for work was the water from Black River. The survey was conducted in May and November. Measuring points from which the sample was taken for analysis were: Measuring point 1 (MM1) Black River near the village Novaci, measuring point 2 (MM2) Black River near the village Kravari, measuring point 3 (MM3) Black River after the junction with Fifth channel and measuring point 4 (MM4) Black River in the village of Skochivir.

Microbiological analysis included examination of the following groups of microorganisms: determination of total number of Coliform bacteria-colimetric method according to Mostprobablenumber by Mac Conkey-Broth at 37° C, total number of Fecal coliform bacteria-colometric method, on liquid substrate Mac Conkey-Broth at 44° C, determination on the number of Escherichia coli-membrane filtration method and determination of the number of Enterococcus-membrane filtration of Slantez Bartley Agar. From the obtained results it can be seen that the water in measuring point 1 from all other measuring points from the hygienic and sanitary aspect is of the best quality and the values for total Coliform bacteria in the water were the lowest with a value of 22000 coliform bacteria/L, Fecal coliform bacteria were 40000 faecal coliform bacteria/L in May. In measuring point 3 was found the largest number of all types of bacteria that were the subject of research.

By summarizing the obtained results, it can be concluded that high values of Coliform bacteria, Fecal coliform bacteria is high due to the presence of wastewater and fecal water that flows from the canal in Black River. Water pollution is closely related to anthropogenic impact and therefore requires constant monitoring, monitoring of water quality changes.

**Key words:** water, microbiological indicators, coliform bacteria, fecal coliform bacteria, escherichia coli, enterococcus, hygienic and sanitary aspect

## Introduction

The increase of the human population, in the world and in our country, the modern way of life, the rapid industrialization as well as the climate change have a significant impact on the deterioration of water quality (Angelevski, 2000; Altinoluk et al., 2014).

The need for clean water imposes the need for constant monitoring of surface and groundwater quality. According to Obradović et al. (2006), carriers of larger amounts of waste materials are surface waters, which are burdened by the action of anthropogenic factor. According to Đukić et al. (2000), the presence of microorganisms in an aquatic system depends on several environmental factors: temperature, solar radiation and its permeability, the availability of dissolved oxygen, the amount of organic matter, the characteristics of the water basin, so that they can all stimulate or inhibit their growth and development.

The purpose of this research is to determine the condition of the Black River, from a hygienic-sanitary aspect.

## Material and Methods

The material used for the analysis was the water from the Black River. The selection of measuring points was at 4 different locations on Black River in the Pelagonija region in the Republic of North Macedonia. The survey was conducted in May and November. Measuring points from which a sample was taken for analysis were: Measuring point 1 (MM1) Black River near the village Novaci. This measuring point is 11km away from the City of Bitola. Measuring point 2 (MM2) before the water flows from the fifth channel in the Black River.

Measuring point 3 (MM3) Black River after the junction with Fifth canal into which the wastewater (industrial and fecal) water flows

and Measuring point 4 (MM4) Black River in the village Skochivir.

In order to determine the water quality from a sanitary-hygienic aspect, the following microbiological tests were performed: determination of the total number of Coliform bacteria-colimetric method according to Mostprobablenumber (MPN) of Mac Conkey - Broth at 37° C for 48 hours (Kungulovski, 2005), determination of Fecal coliform bacteria-colometric method, on liquid substrate Mac Conkey-Broth at 44° C for 24 hours (Kungulovski, 2005), determination of the number of Escherichia coli-method of membrane filtration. The filter is placed on lactose TTS Agar at a temperature of 36° C for 21 hours. Oxidase test and indole test are performed. Incubation is on non-selective agar (TryptoneSoyAgar) at 36° C for 21 hours-tryptophanbroth oxidase test is incubated at 44° C for 21 hours, determining the number of Enterococcus-membrane filtration on special Streptococcal agar (Slantez Bartley Agar). The incubation is at a temperature of 36° C for 44 hours.

## Results and Discussion

Water is an existential and limited natural resource. Due to the uneconomical and irrational use of available water, the amount of polluted water increases, which has a negative impact on the overall environment

Microbiological parameters are one of the most important parameters for water quality. Coliform bacteria are an indicator of water quality and safety and are an indicator for determining water quality (Boron, 2015).

From the results shown in figure 1 it can be concluded that the highest value of total Coliform bacteria is in MM3 2400000 total coliform bacteria/L, which is due to the inflow of the Fifth canal in Black River. At this measuring point, all fecal and industrial wastewater is transferred from

the canal to Black River. The lowest value in MM1 is 22000 total coliform bacteria/L in the spring period - in May. The presence of this group of bacteria in the water indicates a recent introduction of waste of human or animal origin.

If waste and fecal matter are discharged into the recipient, such waters can be a source of pathogenic diseases in humans (Bitton, 2002-a; Blazhevska, 2016). One of the factors on which the number of coliform bacteria of fecal origin depends is the population density.

From figure 2 it can be concluded that the largest number of fecal coliform bacteria is in the river Crna in November at measuring point 3 (MM3 270,000 fecal coliform bacteria/L) The results obtained according to the classification from Kavka (1994) for the degree of fecal pollution, indicate that the water in MM3 is polluted to very polluted. The pronounced large number of fecal coliform bacteria in November is due to the abundant atmospheric sediments which, washing the soil enriched with manure of animal origin, introduce fecal coliform bacteria into the water of Black River.

The lowest number of Fecal coliform bacteria was measured in May in MM1 40000 Fecal coliform bacteria/L, with water according to the Kavka (1994) classification being considered polluted.

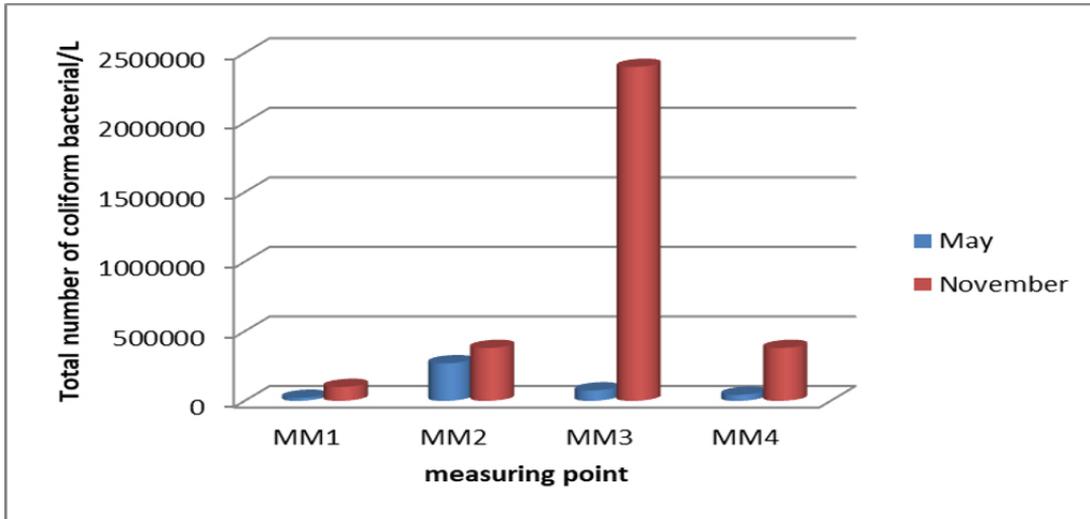
Bacteria of the species *Escherichia coli* are considered as an indicator of the degree of water pollution from a sanitary microbiological aspect (Petrović et al., 1998; Ziberoski 2000; Ziberoski 2001).

From the results shown in figure 3, it can be concluded that *Escherichia coli* is present in all examined samples of water from the river Crna.

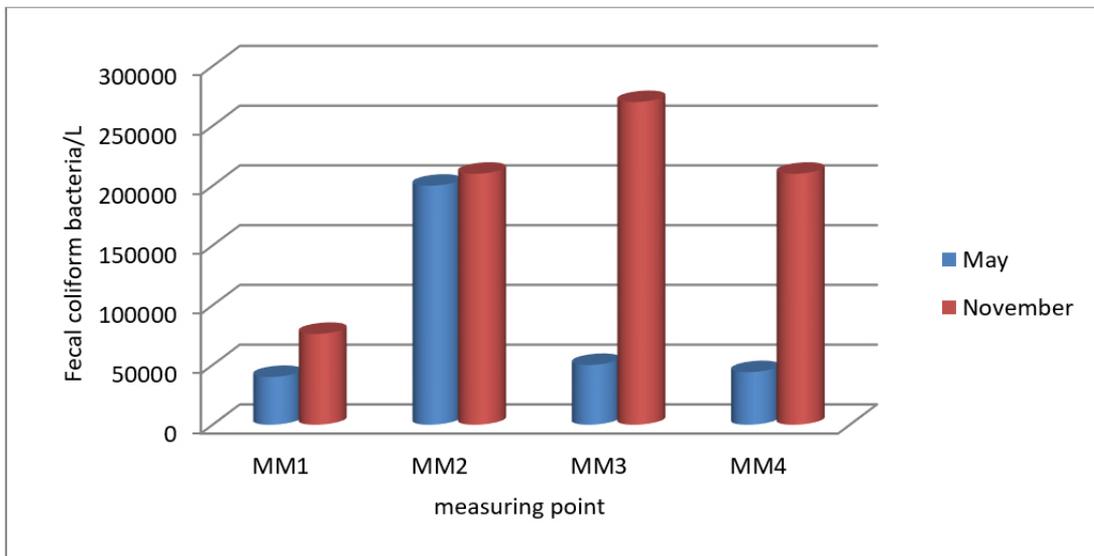
The highest number of *Escherichia coli* in the examined samples was recorded in MM3 200000 *Escherichia coli*/100 mL in November and in MM2 in May. In MM3 the number of Fecal coliform bacteria is the highest. On based to the presence of fecal coliform bacteria, according to Kavka (1994), the water in this measuring point is in V class. The reason for the high value, a part from the anthropogenic impact, is the presence of livestock farms in the area (Cabral et al., 2010; Boron et al., 2015).

Representatives of the genus *Enterococcus* were separated from the genus *Streptococcus* during the 1980s. The genus is formed by relatively different groups that show similar phenotypic characteristics. Most species of this genus originate from the intestinal flora of mammals, birds, reptiles, and humans. Some species have been isolated from the urinary tract as well as from infected wounds (Cabral 2010).

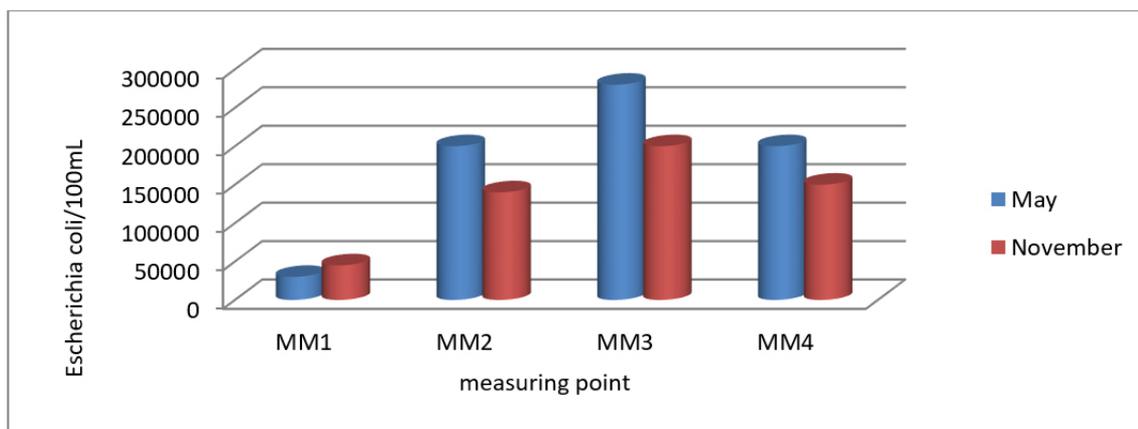
Figure 4 shows the presence of the genus *Enterococcus* at all measuring points in the water of Black River. The highest prevalence of enterococci was observed at MM4 150000 bacteria/100 mL in November, and the lowest prevalence at MM1 30000 bacteria/100 mL in May. Probably the higher prevalence of *Enterococcus* in the waters of Black River, a part from the inflow of water from the Fifth channel which is loaded with fecal wastewater, according to Blazhevska (2016) also have atmospheric sediments (precipitation) with which enterococci are introduced from the soil, i. e. the plant material present along the river. According to the Decree on water classification (No. 18/99), the water in measuring point 1, 2, 3 and 4 is in V class.



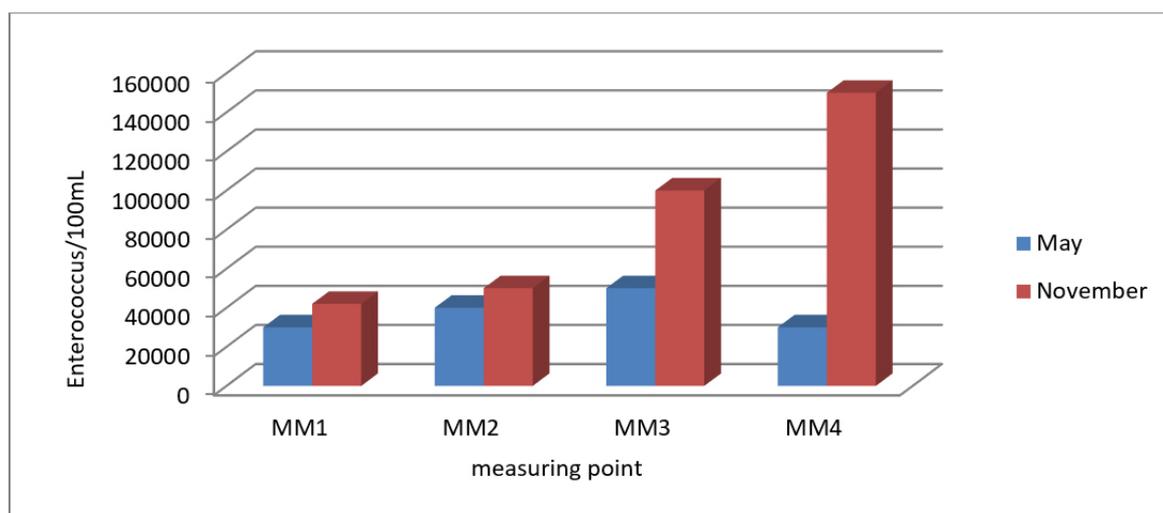
**Fig. 1.** Total Coliform bacteria in the water of Black River in May and November



**Fig. 2.** Fecal coliform bacteria in the water of Black River in May and November



**Fig. 3.** Presence of Escherichia coli in the water of Black River in May and November



**Fig. 4.** Presence of Enterococcus in the water of Black River water in May and November

## Conclusion

Based on the results obtained during the research of water quality in Black River it can be concluded that.

The high values of the total coliform bacteria are characteristic for the settlements where the intake of fecal water from the septic tanks also has an impact.

Fecal coliform bacteria are mostly present in

the water of Black River, in measuring point 3. According to the Decree on classification of surface waters in the Republic of North Macedonia, the water in Black River belongs to the fourth class water, where the number of Fecal bacteria is above 500 in 100/ml water.

Escherichia coli, as a typical representative of coliform bacteria, is present in the water at all measuring points in Black River. The highest presence in the water of Black River is in the

measuring point 3. In MM3 the number of coliform bacteria is the highest due to the presence of wastewater and fecal water that flows from the canal. The reason for the high value, a part from the anthropogenic impact is the presence of livestock farms in the area.

The genus *Enterococcus* is considered a bio-indicator that indicates contamination of water with pathogenic microorganisms. The presence of enterococci was found in all measuring points and with the highest value in MM4. Probably the higher prevalence of *Enterococcus* in the waters of Black River, a part from the inflow of water from the Fifth channel which is loaded with fecal wastewater, according also have atmospheric sediments (precipitation) with which enterococci are introduced from the soil, i.e. the plant material present along the river.

Based on the results of the water quality research in Black River, a general conclusion can be drawn that water pollution is closely related to anthropogenic impacts and therefore requires constant monitoring, monitoring of water quality changes.

## References

**Altinoluk P., Camur-Elipek B., & Aydogduh, H.** (2014). Vertical dynamics of some indicator microorganisms in Tunca river at Turkish Thrace. *Macedonian Journal of Ecology and Environment*, 16, 5-9.

**Angelevski, A.** (2000). Microbiology of water, food and air. NIP Studentski zbor, Skopje.

**Bitton, G.** (2002-a). Encyclopedia of environmental microbiology, Fecal contamination, sources of: 1265/Copyright 2002 by *John Wiley & Sons, Inc.*, New York.

**Blazhevaska, T.** (2016). Microbiological contamination and physicochemical characteristics of the water in the Fifth canal and its impact on the water quality the Black River. Doctoral dissertation. University "St. Cyril and Methodius"- Skopje. Faculty of Agricultural Science and Food – Skopje.

**Boron L. A., Puchala M., & Boron P.** (2015). Potential Microbiological Threat to the Vistula Waters by Its Tributaries in the Vicinity of Kraków, Poland. *Soil & Water Res.*, 130-136.

**Cabral, J.** (2010). Water Microbiology. Bacterial Pathogens and Water. *International Journal of Environmental Research and Public Health*, 1660-4601.

**Đukić, D., Gajin, S., & Matavulj, M.** (2000). Water Microbiology, *Prosveta*, Belgrade, Serbia

**Kavka, G. G. & Poetsch, E.** (1994). Microbiology. Technical Report of the International Commission for the Protection of the Danube River. Eds. Péter Literáthy, Veronika Koller Kreimel, Igor Liska. Eigenverlag ICPDR. 138-150.

**Kungulovski, Dj.** (2005). Microbiology. *University, Ss. Cyril and Methodius* " Skopje.

**Obradović, V., Petković, A., Burger, B., & Petrović, O.** (2006). Experiences, needs and importance of biological analyzes in the control of wastewater treatment plants. *Subotica*.

**Petrović O., Gajin S., Petrović O., Matavulj M., Radnović D., & Svirčev Z.** (1998). Microbiological examination of surface water quality. Institute of Biology. Novi Sad, p. 108. Decree on water classification (1999). Official Gazette of the Republic of Macedonia. Macedonia No.18.

**Ziberovski, J., Naumovski, M., Iljovski, I., & Jankulovski, L.** (2001). The salinity of the water from the river Treska and its use for fish breeding and irrigation. Eighth conference "Water Economy in the Republic of Macedonia", 199-203, Struga.

**Ziberovski, J., Naumovski, M., Iljovski, I., Jankulovski, L., & Hristovska, I.** (2000). Saprophytic characteristics of water from the river. Sticker and possibilities for use for irrigation. Water Economy in the Republic of Macedonia, Seventh conference, Struga.