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## Water quality at special nature reserves in Vojvodina, Serbia

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### ABSTRACT

Although protected, special nature reserves are not isolated from anthropogenic influences entering from outside their borders. Water quality is particularly endangered at three special nature reserves in Vojvodina, i.e. Ludaško jezero (Ludas Lake), Carska bara (Imperial Pond) and Obedska bara (Obed Pond). The special nature reserve Obed Pond and Ludas Lake are polluted by agricultural runoff and untreated wastewater resulting in serious eutrophic processes, and the SNR Imperial Pond is potentially influenced from nearby fish pond and agriculture. The monitoring conducted in summer and in autumn in 2016, at the three mentioned special nature reserves, included water sampling on several locations within each protected area. The analyses included basic water quality parameters, microbiological analyses, as well as water mineralization. Both, water quality and microbiological results, have proven the presence of eutrophic processes at all analyzed protected areas, but the worst conditions were detected in the North and Middle of the Ludas Lake. Considering the other two investigated protected areas, results are a bit better indicating moderate eutrophic conditions.

### Introduction

Special nature reserves are protected because of preserved wildlife, and especially vulnerable are those where water regime is crucial for maintaining their stability. In the Province of Vojvodina, protected moist areas belong to wetlands, ponds and swamp forests, or saline habitats like lakes or salt marshes. The research conducted during 2016 represents an extension of a monitoring done in 2015 (Grabić et al., 2016) aimed at investigating three special nature reserves (SNRs) in Vojvodina: SNR Ludas Lake (Ludaško jezero), SNR Imperial Pond (Carska bara) and SNR Obed Pond (Obedska bara). What is common in all mentioned SNRs is the fact that water plays a major role in defining their characteristics, which are quite different by their origin and features. All SNRs are complex and are comprised of mosaic of natural ecosystems (e.g. SNR Imperial Pond consists of river, pond, swamp, salt marsh, steppe, meadow and forest) and anthropogenic ones (Amidžić et al., 2007). In this research, the focus was on water bodies that are dominating the reserves:

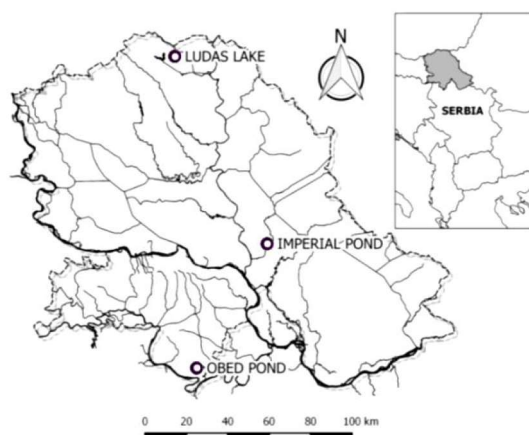
Ludas Lake, Old Bega River within the SNR Imperial Pond and Obed Pond. Whereas SNR Ludas Lake represents a shallow saline lake, Imperial Pond and Obed Pond have originated from river meanders; the first one from the Bega River (reka Begej) and the second one from the Sava River. Both ponds are strongly influenced by the water regime of the rivers. Unlike the mentioned, Ludas Lake was formed by draining the water from the sandy area and a series of salty depressions and from the River Kires (reka Kireš) (Amidžić et al., 2007). With regard to the mentioned diversity and the fact that the SNR were not included in the regular national monitoring, the aim of the research was to examine water quality in order to determine the presence of nutrients, which are promoters of eutrophic processes, as well as major microbiological indicators.

### Materials and methods

The monitoring was conducted in two cycles: the 1<sup>st</sup> was at the end of June/beginning of July and the 2<sup>nd</sup> was at the end of October/beginning of November in 2016 at the three mentioned SNRs, whose location is presented in Fig. 1. Apart from different characteristics and origin,

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the three SNR were chosen because of their relatively distant/dispersed position within the province of Vojvodina (Fig. 1).



**Fig. 1.** Location of the investigated SNRs

SNR Ludas Lake is located in the north of Vojvodina, close to the border with Hungary. SNR Imperial Pond is positioned in the central part of the Banat Region, in the alluvial plane of the Rivers Tisa and Bega, and SNR Obed Pond is situated in the alluvial plane of the River Sava in the very south of the Vojvodina province (Amidžić et al., 2007).

The monitoring conducted in the summer (June-July) and in the autumn period (October-November) in 2016 at all SNRs, included water sampling. The samples were taken from three sampling points at SNR Ludas Lake: North, Middle and South. The sampling conducted at SNR Imperial Pond was conducted at four sampling points: in the areas under protection – Tractor Pond (Traktor bara) and the Old Bega River (Stari Begej) - auxiliary dock; and outside the protected areas at a Sifon canal – a canal buffering influences of commercial fish ponds towards protected areas, as well as from the fish pond. At SNR Obed Pond, sampling was done at two sampling points: Obrez (Obrež) and ameliorative canal Revenice, which flows into the protected pond. All samples were taken from 0 to 50 cm depth, as a grab sample.

Some basic water quality parameters were measured on site (air and water temperature, dissolved oxygen (DO), % of DO saturation, pH and conductivity) and the rest was analyzed in the laboratory of the Faculty of Agriculture, University of Novi Sad. Laboratory analyses included suspended solids (SS), biochemical oxygen demand ( $BOD_5$ ), and nutrient parameters: nitrites ( $NO_2-N$ ), nitrates ( $NO_3-N$ ), orthophosphates ( $PO_4-P$ ), total nitrogen (TN) and total phosphorus (total P). Microbiological analysis included the determination of total number of: saprophytic bacteria, fungi, coliform

bacteria, *Escherichia coli* and the number of sulphite reducing clostridia (genus *Clostridia*). The number of microorganisms was determined using the dilution method (Trolldenier, 1996). Appropriate nutrient media were used: nutrient agar for the number of saprophytic bacteria, Chapeck–Dox agar for fungi, Hi Crome Salmonella agar for the number of coliform bacteria and *Echerichia coli*, and medium with pepton (pepton 15 g/L, extract yeast 9 g/L,  $Na_2SO_3$  0.5 g/L, agar 15 g/L; after medium sterilization, 20 ml of 7% solution of  $FeSO_4$  was added) for the sulphite reducing clostridia.

## Results and discussions

### Biochemical water quality

The results of the monitoring, conducted at three SNRs in two cycles (summer and autumn) in 2016, are shown in Tables 1-3.

Chemical water quality status at investigated sampling points at SNR Ludas Lake may be characterized as poor - bad (IV-V water class), for all sampling points and both cycles, except for the sampling point South during the autumn, where moderate status (III water class) was registered (Table 1). The classification was done according to Serbian national regulations, which are in line with the EU Water Framework Directive (Regulation 74/2011/RS; Regulation 50/2012/RS). Generally, at all sampling points, eutrophic processes, i.e. 'water bloom', were present.

The processes are obvious, especially in the North and Middle sampling point, where green colour of the water and extremely high concentrations of DO and % DO saturation are indicating supersaturation in most cases. Values of DO parameters coupled with high values of pH,  $BOD_5$  and nutrients (TP and TN) are proving evidence for the abundance of phytoplankton. Apart from the elevated concentrations of nutrients, which is gradually decreasing from the north to the south of the lake, there is also gradual, but opposite change in water salinity (Rudić et al., 2014; Petrović, 1980). The research also confirmed this phenomenon, i.e. an increase of electrical conductivity from the North to the South sampling point, which is due to the natural leaching of salts from adjacent salty depressions (Amidžić et al., 2007). High values of the electric conductivity, DO, % DO saturation, the content of nutrients and high pH are in accordance with the results obtained in 2015 (Grabić et al., 2016), as well as by the other authors (Rudić et al., 2014). Low values of inorganic compounds  $NO_2-N$ ,  $NO_3-N$  and  $PO_4-P$  and relatively high values of TN and TP are indicating that majority of nutrients is present in an organic form in water.

**Table 1.** Water quality at SNR LudasLake in 2016

| No. | Sampling points:   |                      | 1. North |        | 2. Middle |        | 3. South |        | Limit values set by the regulations (Regulation 74/2011/RS; Regulation 50/2012/RS) |                    |                    |                    |
|-----|--------------------|----------------------|----------|--------|-----------|--------|----------|--------|--|--------------------|--------------------|--------------------|
|     | Parameters:        | Date:                | 21.07.   | 04.11. | 21.07.    | 04.11. | 21.07.   | 04.11. | I  | II                 | III                | IV                 |
|     |                    | Units:               |          |        |           |        |          |        |  |                    |                    |                    |
| 1.  | T <sub>air</sub>   | °C                   | 26.6     | 12     | 26.6      | 10     | 27.2     | 10     | -  | -                  | -                  | -                  |
| 2.  | T <sub>water</sub> | °C                   | 24.1     | 10     | 24.5      | 8.2    | 26.2     | 8.1    | -  | -                  | -                  | -                  |
| 3.  | DO                 | mg O <sub>2</sub> /L | >17      | 15.09  | 13.26     | 11.68  | 15.35    | 9.95   | 8.5*   | 7.0                | 5.0                | 4.0                |
| 4.  | % DO sat.**        | %                    | >217     | 135    | 161       | 99     | 195      | 85     | 70-90  | 50-70<br>(105-115) | 30-50<br>(115-125) | 10-30<br>(125-130) |
| 5.  | pH                 | -                    | 10.0     | 8.66   | 9.28      | 8.73   | 9.1      | 8.0    | 6.5 - 8.5  |                    |                    | <6.5;<br>>8.5      |
| 6.  | Cond.              | µS/cm                | 911      | 936    | 1245      | 1271   | 1795     | 1616   | <1000*   | 1000               | 1500               | 3000               |
| 7.  | SS                 | mg/L                 | 79       | 53     | 36        | 18     | 12       | 10     | 25   | 25                 | -                  | -                  |
| 8.  | BOD <sub>5</sub>   | mg O <sub>2</sub> /L | 23.5     | 9.8    | 23.0      | 5.4    | 15.8     | 4.3    | 1.5  | 5.0                | 6.0                | 20.0               |
| 9.  | NO <sub>2</sub> -N | mg N/L               | <0.01    | <0.01  | <0.01     | <0.01  | 0.01     | 0.03   | 0.01*  | 0.03               | 0.12               | 0.3                |
| 10. | NO <sub>3</sub> -N | mg N/L               | <1       | <1     | <1        | <1     | 2.7      | <1     | 1.50   | 3.00               | 6.00               | 15.00              |
| 11. | TN                 | mg N/L               | 1.7      | 2.6    | 1.2       | 2.6    | <0.5     | 2.9    | 1*   | 2                  | 8                  | 15                 |
| 12. | PO <sub>4</sub> -P | mg P/L               | <0.02    | 0.26   | 0.09      | 0.06   | 0.06     | 0.14   | 0.02   | 0.1                | 0.2                | 0.5                |
| 13. | TP                 | mg P/L               | 0.48     | 0.13   | 0.32      | 0.06   | 0.30     | 0.17   | 0.05   | 0.2                | 0.4                | 1.0                |

\*or natural level; \*\*limit values for supersaturation are given according to the Regulation 2/74 and 24/76/SFRY

**Table 2.** Water quality at SNR Imperial Pond in 2016

| No. | Sampling points:   |                      | 1. Tractor Pond |        | 2. Old Bega River |        | 3. Sifon canal | 4. Fish Pond |        | Limit values set by the regulations (Regulation 74/2011/RS; Regulation 50/2012/RS) |                    |                    |                    |
|-----|--------------------|----------------------|-----------------|--------|-------------------|--------|----------------|--------------|--------|--|--------------------|--------------------|--------------------|
|     | Parameters:        | Date:                | 7.7.            | 28.10. | 7.7.              | 28.10. | 28.10.         | 7.7.         | 28.10. | I  | II                 | III                | IV                 |
|     |                    | Units:               |                 |        |                   |        |                |              |        |  |                    |                    |                    |
| 1.  | T <sub>air</sub>   | °C                   | 25.6            | 11.6   | 25.0              | 11.0   | 8.6            | 27.7         | 11.6   | -  | -                  | -                  | -                  |
| 2.  | T <sub>water</sub> | °C                   | 25.6            | 11.8   | 23.3              | 10.6   | 11.0           | 25.4         | 10.9   | -  | -                  | -                  | -                  |
| 3.  | DO                 | mg O <sub>2</sub> /L | 5.9             | 11.64  | 3.9               | 10.27  | 11.00          | 3.36         | 11.51  | 8.5*   | 7.0                | 5.0                | 4.0                |
| 4.  | % DO sat.**        | %                    | 72.7            | 107.8  | 67.4              | 92.6   | 98.8           | 41.3         | 104.2  | 70-90  | 50-70<br>(105-115) | 30-50<br>(115-125) | 10-30<br>(125-130) |
| 5.  | pH                 | -                    | 8.00            | 7.97   | 7.45              | 7.77   | 7.85           | 7.50         | 8.22   | 6.5 - 8.5  |                    |                    | <6.5;<br>>8.5      |
| 6.  | Cond.              | µS/cm                | 355             | 454    | 570               | 515    | 518            | 526          | 565    | <1000*   | 1000               | 1500               | 3000               |
| 7.  | SS                 | mg/L                 | 19              | 9      | 21                | 71     | 7              | 42           | 54     | 25   | 25                 | -                  | -                  |
| 8.  | BOD <sub>5</sub>   | mg O <sub>2</sub> /L | -               | 0.0    | 9.8               | 0.0    | 0.0            | 18.6         | 6.5    | 1.5  | 5.0                | 6.0                | 20.0               |
| 9.  | NO <sub>2</sub> -N | mg N/L               | <0.01           | <0.01  | <0.01             | 0.02   | <0.01          | <0.01        | <0.01  | 0.01*  | 0.03               | 0.12               | 0.3                |
| 10. | NO <sub>3</sub> -N | mg N/L               | <1              | <1     | <1                | <1     | <1             | <1           | <1     | 1.50   | 3.00               | 6.00               | 15.00              |
| 11. | TN                 | mg N/L               | 2.2             | 2.7    | 2.9               | 4.0    | <0.5           | 2.0          | 1.5    | 1*   | 2                  | 8                  | 15                 |
| 12. | PO <sub>4</sub> -P | mg P/L               | 0.07            | <0.02  | 0.10              | <0.02  | 0.06           | 0.20         | <0.02  | 0.02   | 0.1                | 0.2                | 0.5                |
| 13. | TP                 | mg P/L               | 0.33            | 0.13   | 0.38              | 0.16   | 0.06           | 0.59         | 0.30   | 0.05   | 0.2                | 0.4                | 1.0                |

\*or natural level; \*\*limit values for supersaturation are given according to the Regulation 2/74 and 24/76/SFRY

Water quality in SNR Imperial Pond at the sampling points, within and outside the SNR, may be characterized as moderate to poor (III-IV water class), if the parameter, whose value indicates the worst class, is the one which determines water quality class (Table 2). Generally, parameters which were of concern, were oxygen parameters, BOD<sub>5</sub>, SS and total P. Especially low concentrations of DO and % DO saturation were measured in July at sampling points under the protected area, i.e. Tractor Pond and

Old Bega River, and Fish Pond, which is managed in a way to primarily support fish production. A common feature is that all mentioned water bodies, where sampling was done, belong to stagnant or slow flowing water. The only watercourse – Sifon canal, although representing anthropogenic creation, brings fresh water from the Tisa River to the SNR's area, contrary to the Old Bega River, which passes through the protected area, where water is predominantly stagnant.

**Table 3.** Water quality at SNR Obed Pond in 2016

| No. | Sampling points :  |                      | 1. Obrez |        | 2. Revenice |        | Limit values set by the regulations<br>(Regulation 74/2011/RS; Regulation 50/2012/RS) |                    |                    |                    |
|-----|--------------------|----------------------|----------|--------|-------------|--------|---|--------------------|--------------------|--------------------|
|     | Parameters:        | Date:                | 24.6.    | 20.10. | 24.6.       | 20.10. | I   | II                 | III                | IV                 |
|     |                    | Units:               |          |        |             |        |   |                    |                    |                    |
| 1.  | T <sub>air</sub>   | °C                   | 31.2     | 17.1   | 31.7        | 17.1   | -   | -                  | -                  | -                  |
| 2.  | T <sub>water</sub> | °C                   | 27.4     | 12.3   | 25.0        | 10.9   | -   | -                  | -                  | -                  |
| 3.  | DO                 | mg O <sub>2</sub> /L | 8.00     | 6.28   | 1.66        | 6.22   | 8.5*  | 7.0                | 5.0                | 4.0                |
| 4.  | % DO sat.**        | %                    | 103      | 58.4   | 20.44       | 56.40  | 70-90   | 50-70<br>(105-115) | 30-50<br>(115-125) | 10-30<br>(125-130) |
| 5.  | pH                 | -                    | 7.54     | 7.76   | 7.17        | 7.61   | 6.5 - 8.5   |                    |                    | <6.5;<br>>8.5      |
| 6.  | Cond.              | µS/cm                | 469      | 505    | 469         | 501    | <1000*  | 1000               | 1500               | 3000               |
| 7.  | SS                 | mg/L                 | 7        | 12     | 10          | 12     | 25  | 25                 | -                  | -                  |
| 8.  | BOD <sub>5</sub>   | mg O <sub>2</sub> /L | 32.3     | 3.8    | 37.3        | 2.7    | 1.5   | 5.0                | 6.0                | 20.0               |
| 9.  | NO <sub>2</sub> -N | mg N/L               | 0.02     | <0.01  | 0.02        | 0.02   | 0.01*   | 0.03               | 0.12               | 0.3                |
| 10. | NO <sub>3</sub> -N | mg N/L               | <1       | <1     | <1          | <1     | 1.50  | 3.00               | 6.00               | 15.00              |
| 11. | TN                 | mg N/L               | 1.9      | 1.6    | 2.3         | 1.8    | 1*  | 2                  | 8                  | 15                 |
| 12. | PO <sub>4</sub> -P | mg P/L               | 0.44     | 0.1    | 0.37        | 0.13   | 0.02  | 0.1                | 0.2                | 0.5                |
| 13. | TP                 | mg P/L               | 0.42     | 0.02   | 0.32        | 0.07   | 0.05  | 0.2                | 0.4                | 1.0                |

\*or natural level; \*\*limit values for supersaturation are given according to the Regulation 2/74 and 24/76/SFRY

Results of monitoring at the SNR Obed Pond indicate that water quality is good to poor (II-IV water class) (Table 3). Parameters that are responsible for such characterization are oxygen parameters, BOD<sub>5</sub>, total TN and TP.

#### Microbiological water quality

At all three SNRs, in both cycles (summer and autumn), there was a high number of saprophytic bacteria ranging from 950 bacterial cells/mL of water registered in June at sampling points of SNR Obed Pond, to  $9.67 \times 10^4$  cells/mL in water of SNR Ludas Lake - sampling point North in November 2016. Saprophytic mesophilic bacteria have an important role in the process of water purification. It is interesting that these bacteria can use many pollutants as sources of nutrients and energy for their metabolism.

Fungi belong to the exclusive and intensive aerobic decomposers of organic matter (Gleason et al., 2018). In the waters of the investigated areas, the number of this group of microorganisms was very low (max 38 cells/mL in October at the sampling point Tractor Pond - SNR Imperial Pond). Reduced numbers of fungi may indicate lack of oxygen in the tested water.

The total number of sulphite-reducing clostridia in water at the sampling points was low and did not exceed the limit of  $10^3$  cells/mL, which indicates the existence of anaerobic conditions in the water and a substantial lack of oxygen. The highest

number was registered at the sampling point Old Bega River within SNR Imperial Pond in July 2016 and amounted to 175 cells/mL.

Coliform bacteria are the most desirable group of indicators of faecal pollution of water. They are common bacteria in the environment and are generally not harmful. These are excreted by faeces, and afterwards, through wastewaters, they reach natural recipients – water bodies. If the natural water is loaded with faeces, then parasites, pathogenic bacteria and viruses are likely to be present (Costán-Longares et al., 2008; Oana et al., 2017). Potentially dangerous place in the context of this research was the sampling point North (SNR Ludas Lake), where, in the autumn of 2016,  $3.65 \times 10^4$  cells of coliform bacteria per mL of water were found. The purest water was detected at SNR Obed Pond in the summer of 2016, at both sampling points, which may be due to previous rainy period.

*Escherichia coli* is a cause of many diseases such as diarrhoea, infections, urinary tract infections, sepsis, and others. In addition, the bacterium serves as an indicator of water quality. The presence of *Enterobacteriaceae E. coli* in water at all sampling points, in the spring/summer and autumn in 2016 was very low, which suggests that, despite the high number of bacteria, waters of SNRs Ludas Lake, Imperial Pond, and Obed Pond have a self-cleaning capacity. Figures 2-4 show the results of microbiological analyses.

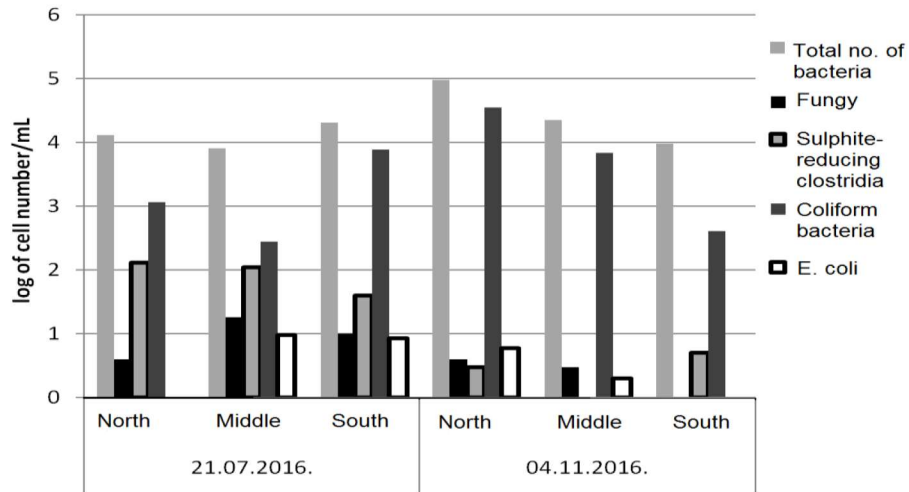


Fig. 2. Number of the examined groups of microorganisms at SNR Ludas Lake in 2016

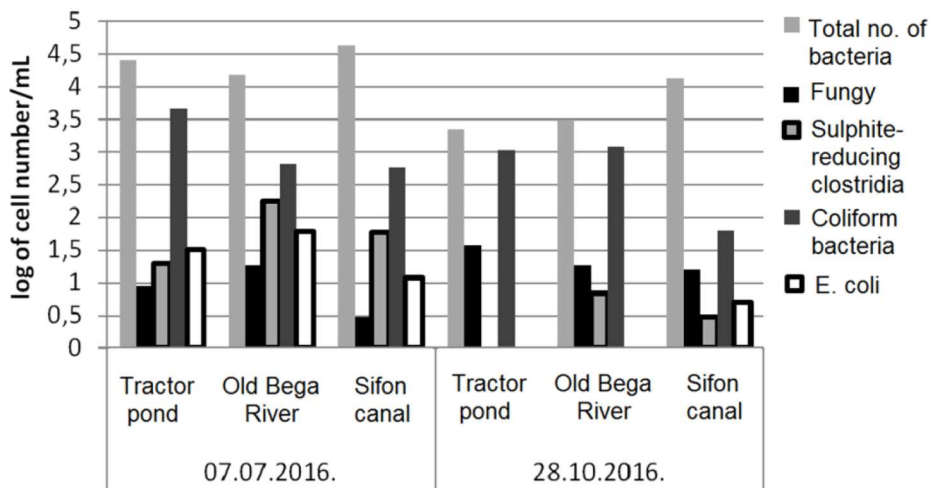


Fig. 3. Number of the examined groups of microorganisms at SNR Imperial Pond in 2016

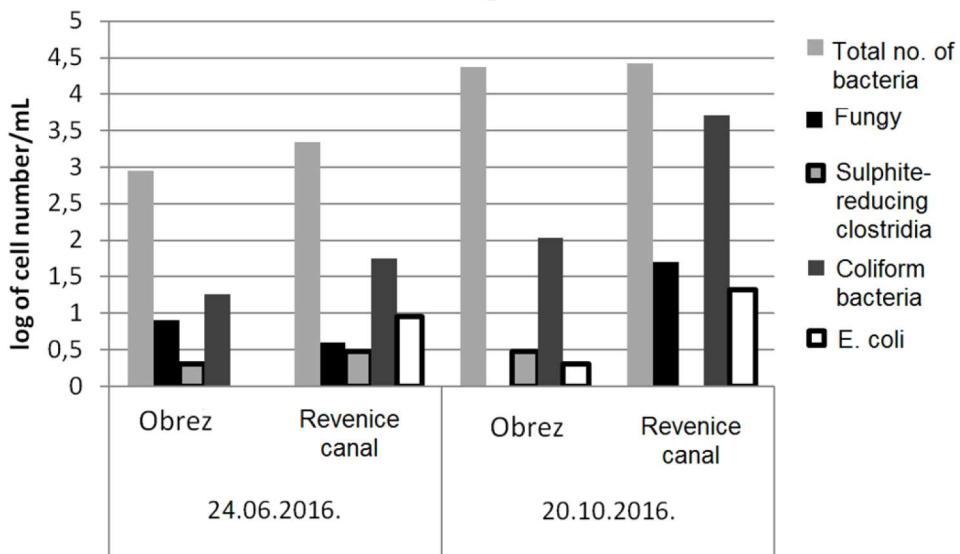


Fig. 4. Number of the examined groups of microorganisms at SNR Obed Pond in 2016

## Conclusions

The analyses of both biochemical and microbiological water quality indicators, in 2016, have revealed the presence of eutrophic processes at all sampling points. Both sets of analyses have proven high eutrophic status of the SNR Ludas Lake and significant salinity and differences between north and south part of the lake, which is due to a natural saline character of the lake and its surrounding. The results of water quality of the other two investigated SNRs are a bit better indicating moderate eutrophic conditions. Due to the abundance of different groups of microorganisms, it can be concluded that sampling point North, within the SNR Ludas Lake, had greater eutrophication compared to all other sampling points at investigated SNRs.

The monitoring needs to be continued in the following years in order to obtain a clear picture of the extent and changes of eutrophic processes, which are significantly dependent on variable climatic conditions and anthropogenic influence. It is also necessary to continue water quality monitoring of all inflows of watercourses which bring certain pollution load to the protected areas. The obtained result could help managers of SNRs to improve practices of managing water regime within the SNRs, as well as to indicate the necessity of implementing some measures in order to improve water quality of watercourses that are entering protected areas.

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