

## SEASONAL VARIATION OF THE WATER COLOR FROM THE IOR LAKE - BUCHAREST

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**Abstract.** The IOR Lake is located in the eastern part of Bucharest, in the Alexandru Ioan Cuza Park of the 3<sup>rd</sup> district and, based on the available data, is considered a lake of artificial origin, anthropogenic, resulting from the extraction of construction materials from its current perimeter. In relation to the water supply sources, they are included in the category of lakes fed by precipitation (rains, melting snow) but also through some underground springs, having a variable depth. The quality of the lake water is influenced by the organic and inorganic material leached from the slopes that border it, the organic matter accumulated in the sediment, the vegetation that develops on the shores and the presence of economic units (terraces, restaurants, food outlets, etc.). Between October 2020 and April 2021, the colour of the lake water presented a brickish appearance throughout the table, which decreases in intensity in small volumes of water (1-2 litres). The purpose of the obtained analyses and results, as presented in this paper, was to establish a possible cause of this phenomenon. The obtained results showed the presence of mesophilic aerobic heterotrophic bacteria. Of all coliforms in all water samples, colonies characterized by the presence of *E. coli* bacterium, of fecal streptococci and the absence of total coliforms are highlighted. Colonies of ferrobacteria have been identified and from a chemical point of view the presence of iron oxides in larger quantities is noticeable. Physico-chemical data were within normal values.

**Keywords:** IOR Lake, watercolor, water pollution.

**Rezumat. Variația sezonieră a culorii apei din Lacul IOR – București.** Lacul IOR este localizat în partea estică a municipiului București, în parcul Alexandru Ioan Cuza, iar în raport cu datele din literatura de specialitate este considerat un lac de origine antropică, rezultat în urma extragerii materialelor pentru construcții din perimetrul său actual. În raport cu sursele de alimentare cu apă, este încadrat în categoria lacurilor alimentate din precipitații (ploi, topirea zăpezilor), dar și prin intermediul unor izvoare subterane, având o adâncime variabilă. Calitatea apei lacului este influențată de materialul de natură organică și anorganică levigat din versanții care îl mărginesc, materia organică acumulată în sediment, vegetația care se dezvoltă pe maluri și prezența unor unități de natură economică (terase, restaurante, puncte de vânzare produse alimentare etc.). În perioada octombrie 2020 – aprilie 2021, culoarea apei lacului a prezentat în toată masa un aspect cărămiziu, care se diminuează ca intensitate în volume mici de apă (1 – 2 litri). Scopul analizelor a fost acela de a stabili o posibilă cauză a acestui fenomen. Rezultatele obținute au arătat prezența, în toate probele de apă, a bacteriilor heterotrofe aerobe mezofile, a streptococilor fecali și a coliformilor totali. Au fost identificate colonii de ferobacterii, iar din punct de vedere chimic se remarcă prezența oxizilor de fier în cantități mai mari. Datele fizico-chimice s-au încadrat în valori normale.

**Cuvinte cheie:** Lacul IOR, culoarea apei, ape poluate.

### INTRODUCTION

As an essential element of life, water is found on Earth both in liquid form (rivers, lakes, seas, oceans) and in solid form (glaciers). In the atmosphere, water is found in the form of vapors. Human communities are usually developed in the vicinity of water resources, and use them for economic, recreational purposes but also in everyday life. Thus, urban water resources (natural or artificial lakes) are of particular importance due to the large-scale exposure to various pollutants but also as a result of their role in the functioning of metropolitan ecosystems. From this point of view, they are a topic of interest for numerous studies that try to model the temporal evolution but also the predictability regarding their quality and especially the anthropic impact on it (ZHU, 2008; CASTRO-FRESNO et. al., 2009; CHEVERESAN et. al., 2009; TABATABAEI, 2010).

In this context, monitoring the quality of water resources is important both from an economic perspective and from the protection of the environment. Usually, this aspect is achieved by microbiological analyses but also by means of physico-chemical parameters such as temperature, turbidity, pH value, conductivity, etc.

The IOR Lake is located in the eastern part of Bucharest, in the Alexandru Ioan Cuza Park in district 3, and, based on the available data (GHERVASE et. al., 2011) is considered a lake of artificial origin, anthropogenic, resulting from the extraction of construction materials from its current perimeter. Concerning the water supply sources, it is included in the category of lakes fed by precipitation (rains, melting snow) but also through some underground springs, having a variable depth (GHERVASE et. al., 2011).

The quality of the lake water is influenced by the organic and inorganic material leached from the slopes that border it, the organic matter accumulated in the sediment, the vegetation that develops on the shores, and the presence of economic units (terraces, restaurants, food outlets, etc.).

Previous measurements (GHERVASE et. al., 2011) of some physico-chemical parameters showed values of 7.83 mg / L for dissolved oxygen, 83% saturation of water in oxygen, a pH value of 7.82 units, and a conductivity of 981  $\mu\text{S}/\text{cm}^2$ . Thus, cumulating these values with those for nutrients, the literature (GHERVASE et. al., 2011) classifies lake water in classes ranging from II to IV, the general quality of lake water corresponding to class IV compared to

current standards in force (\*\* OFFICIAL MONITOR 161/2006). This classification can be supported by the high ammonium intake but also by the result of laboratory analyses on chemical oxygen consumption that is high (GHERVASE et. al., 2011).

The purpose of this study was to explore the range of factors that can influence water colour change. Chemical composition and bacterial counts (mesophilic aerobic heterotrophic bacteria, total coliforms, faecal coliforms, faecal streptococci, and ferrobacteria) were investigated in the water samples from the IOR Lake.

## MATERIAL AND METHODS

The determination of the physico-chemical parameters of the water samples was performed using the Hanna 9828 multiparameter field.

The absorption spectrum in the visible range was performed using a Specord 210 Plus UV-VIS spectrophotometer.

XRF analysis (X-ray fluorescence spectrometry) of water samples was performed using WD-XRF equipment (Rigaku, Japan), and allowed the identification, as a percentage, of the chemical elements.

The bacteriological analysis of the water samples taken from the IOR Lake (on 12.03.2021) consisted in the use of current analysis methods, according to STAS 3001/1991 and protocols described previously (LUCACI et. al., 2019).

To establish the presence of microbiological indicators of water quality, three water samples were analysed (two – noted 1 and 2 - taken from IBB researchers - sampling on the water column - and one sample, noted 3, collected by the beneficiary, namely National Romanian Water Administration - NRW).

The quantitative determination of the types of bacteria was performed according to the microbiological norms that imply the inoculation of the sample or its dilutions, the incubation, and the determination of the total number of bacteria. Thus, serial decimal dilutions were performed from the water samples, which in the case of aerobic heterotrophic bacteria and total coliforms were determined, inoculated into Petri dishes (in two repetitions) over which solidified growth medium was added. In the case of other groups of bacteria, water samples or appropriate dilutions were inoculated, in three repetitions, into test tubes containing liquid culture media or semi-solid medium was added over the inoculum.

### *Determination of the probable number of mesophilic aerobic heterotrophic bacteria*

The presence of mesophilic aerobic heterotrophic bacteria was highlighted by inoculation of samples and decimal dilutions, by the pour plate method and incubated for 48 hours at 37°C on a nutrient agar medium. Colonies developed on the culture medium were quantified, and the result was expressed by the number of colony-forming units per mL (CFU/mL<sup>1</sup>).

### *Determination of the probable number of total coliforms*

In microbiological analysis, the pour plate technique was conducted in the water samples to find out total coliforms. After 24 hours from incubation at 37°C, on Eosin Methylene Blue Agar culture medium, the number of colony-forming units per mL (CFU/mL<sup>1</sup>) was determined. The presence of total coliforms (Table 1) was confirmed by the development on the culture medium of the characteristic colonies: flat, blue-purple, colonies with metallic lustre, convex or pink colonies, mucoid with dark purple centre.

### *Determination of the probable number of thermotolerant coliform bacteria (faecal coliforms)*

The faecal coliforms were detected in terms of most probable number (MPN) by inoculating the samples and their decimal dilutions, in triplicate, into tubes, over which the bile lactose bromocresol purple culture medium is added, in a high column (MacConkey). The inoculated tubes were incubated at 44-45°C, for 24 hours. The reaction is considered positive when the purple changes to yellow. Taking into account the number of positive tubes, the probable number of microorganisms per mL can be estimated, based on McCrady tables (LAZĂR et al., 2004).

### *Determination of the probable number of faecal streptococci*

The presence of faecal streptococci was highlighted by inoculating in triplicate, in tubes, the selective culture medium (sodium azide broth and purple bromocresol) of the water samples and their decimal dilutions. The seeded tubes were incubated for 48 hours at 44-45°C. Based on the number of confirmed positive tubes (turning purple to yellow), the probable number of microorganisms in one mL of water was determined using the MPN technique.

### *Determination of the probable number of ferrobacteria*

Winogradsky's selective culture medium was prepared for the identification and growth of ferrobacteria. The sample and its dilutions were inoculated by the pour plate method and incubated at 28°C for 7 days. Colonies developed on the culture medium were quantified, and the result was expressed by the number of colony-forming units per mL (CFU/mL<sup>1</sup>). The tests were performed in duplicate (LAZĂR et al., 2004).

## RESULTS

Between October 2020 and April 2021, the colour of the lake water presented a brickish appearance throughout the table (Photos 1 and 2), which decreases in intensity in small volumes of water (1-2 litres). The purpose of the obtained analyses and results, as presented below, was to establish a possible cause of this phenomenon.

The physico-chemical parameters of the water samples are presented in Table 1, and in terms of average values for dissolved oxygen, oxygen saturation, pH value, and conductivity, they are similar to those in literature (GHERVASE et. al., 2011) corresponding thus to the general water quality of the lake with classification in class IV (\*\*\*. OFFICIAL MONITOR 161/2006).



Photo 1. Detail regarding the appearance (colour) of the lake at the sampling points (original).

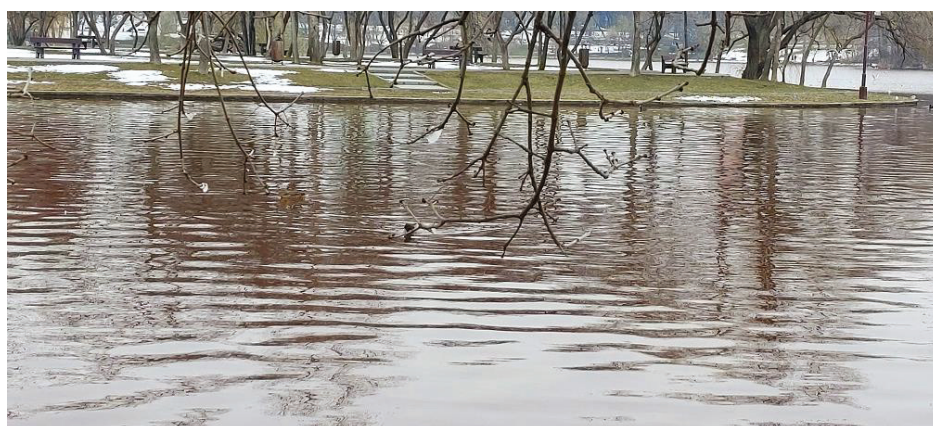


Photo 2. General appearance of the lake at the time of sampling (original, March 12, 2021).

Table 1. The physico-chemical parameters of the investigated samples;  
\* the distance from the bridge to the sampling point was between 10 - 15 m.

Parameters	Sample 1 (right Liviu Rebreanu bridge direction to the Titan metro station) *	Sample 2 (left Liviu Rebreanu bridge direction to the Titan metro station) *
Depth (m)	1.5	1.5
Turbidity	26.7	23
Dissolved oxygen (ppm)	10.25	8.44
pH	6.93	7.97
pH (mV)	-26.8	-82
Temperature (°C)	6.19	6.48
Pressure (mbar)	994.6	992.4
Resistivity (MΩcm)	0.001	0.001
Conductivity 1 (μS/cm)	974	1038
Conductivity 2 (μS/cm <sup>2</sup> )	626	672
Total dissolved solids (ppm)	487	519
Salinity	0.48	0.52
Oxidative-reducing potential	71.9	19.1
Dissolved oxygen saturation (%)	84.5	70.3

The absorption spectrum in the visible range showed a peak with a maximum wavelength of 510 nm. Based on data from the literature (YATSUNAMI et.al., 2014; SELVAM et al., 2015; COJOC et. al., 2019; NEAGU et. al., 2019) this field of absorption corresponds to carotenoid pigments, respectively those in the category of phytoenes and lycopenes, unsaturated compounds with over 40 carbon atoms in the structure.

The XRF analysis of the water sample taken from IOR Lake was performed using WD-XRF equipment (Rigaku, Japan) and allowed the identification of the chemical elements as percentages. For this, a known volume of

each water sample was filtered through filter paper, and the filtrate and filter paper were tested to determine the composition of the chemical elements (Table 2). The results revealed the presence of a significant amount of iron which can support the identification of ferrobacteria and argued for the colour of the water lake.

Table 2. The chemical composition of the investigated sample expressed as mass% from the number of analysed samples; \* the volume of liquid resulting after filtering the sample; \*\* the residue left on the filter after filtering the water; blank cells – no data recorded.

Mass%	Sample 1 – filtered*	Sample 1 – filter**	Sample 2 – filtered*	Sample 2 – filter**	Sample 3 – filtered*	Sample 3 – filter**
Al <sub>2</sub> O <sub>3</sub>	3.99		2.64	3.59		2.62
SiO <sub>2</sub>		2.77	12.96	7.22	12.1	5.06
K <sub>2</sub> O	8.27		6.03		11.49	
CaO	7.91		7.49		9.75	
Fe <sub>2</sub> O <sub>3</sub>		3.31	9.7	8.53		9.38
Pm <sub>2</sub> O <sub>3</sub>	20.03					
Cr <sub>2</sub> O <sub>3</sub>		3.83	6.68			6.38
V <sub>2</sub> O <sub>5</sub>				55.6	25.46	
Ga <sub>2</sub> O <sub>3</sub>	4.33		3.46		5.2	
ZnO			3.08			
NiO		1.28	4.76	1.24	4.48	
TiO <sub>2</sub>			25.28			
Tm <sub>2</sub> O <sub>3</sub>			8.09	7.4		
Eu <sub>2</sub> O <sub>3</sub>	12.44					
Tb <sub>2</sub> O <sub>7</sub>	14.92					
Ho <sub>2</sub> O <sub>3</sub>	6.72					
Co <sub>2</sub> O <sub>3</sub>				1.94		
ZrO <sub>2</sub>				2.07		
Ag <sub>2</sub> O				6.69		19.97
CuO		1.45				
CeO <sub>2</sub>						26.82
MnO					12.75	
ReO <sub>2</sub>					12.17	
MoO <sub>3</sub>						3.53
PbO						4.43
ThO <sub>2</sub>						3.36
GeO <sub>2</sub>						2.7
Rb <sub>2</sub> O						1.65
SrO						1.14
Y <sub>2</sub> O <sub>3</sub>						8.64

The data of the bacteriological analysis showed the presence of the following groups of microorganisms: mesophilic heterotrophic aerobic bacteria (Table 3; Fig. 1), faecal streptococci (Table 3; Fig. 2), total coliforms (Table 3), and ferrobacteria (Table 3; Fig. 3).

Table 3. Quantitative estimation of mesophilic heterotrophic aerobic bacteria, the probable number of total coliforms, the probable number of faecal coliforms, the probable number of faecal streptococci, and the probable number of ferrobacteria.

Sample	mesophilic heterotrophic aerobic bacteria (UFC/mL)	probable number of total coliforms (UFC/mL)	probable number of fecal coliforms (MPN)	probable number of fecal streptococci (MPN)	probable number of ferrobacteria (UFC/mL)
1	5,9x10 <sup>2</sup>	2	0	9	3,2x10
2	2,8x10 <sup>2</sup>	2	0	9	3,3x10
3	8,1x10	2	0	1,8x10	0

Mesophilic heterotrophic aerobic bacteria were determined in all water samples collected from IOR Lake, in numbers of 5.9x10<sup>2</sup> CFU/mL<sup>1</sup>, 2.8x10<sup>2</sup> CFU/mL<sup>1</sup>, and 8.1x10 CFU/mL<sup>1</sup>, respectively. These data showed a numerical abundance of heterotrophic bacteria compared to other groups of bacteria and can be explained by the presence of organic matter that allows the growth and multiplication of organotrophic microorganisms.

The obtained results indicated the presence of total coliforms from the *Enterobacteriaceae* family (*Escherichia coli*) and faecal streptococci in all the analysed water samples. As it is known, faecal and total coliform bacteria are used as indicators of water quality, and their absence from selective culture media leads to a good water quality. In contrast, faecal streptococci, although providing only additional evidence of faecal pollution, are considered better indicators than coliforms because they do not grow and multiply in virgin soils or waters (Selvam et al., 2015). If the ratio between the number of faecal coliform bacteria and the number of faecal streptococci is calculated, the origin of water pollution can be established. Values higher than 1 indicate water pollution with human manure, and values lower than 1 show pollution with animal manure. However, if the ratio between the number of *E. coli* bacteria and the number of faecal streptococci is calculated, then the origin of the pollution should be better defined. In our study, this ratio is less than 1, which shows that water sources are contaminated with animal manure.



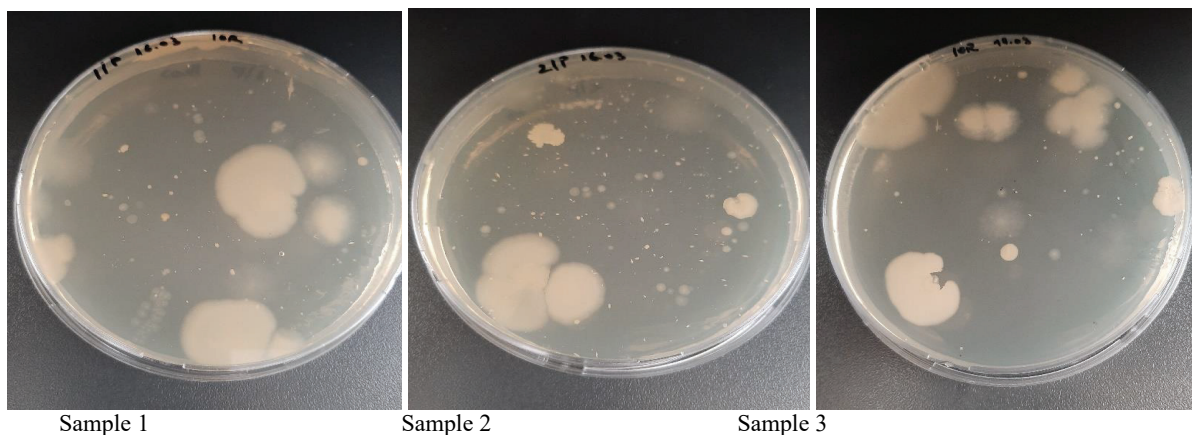


Figure 1. Heterotrophic bacteria isolated from water samples on nutrient agar medium.

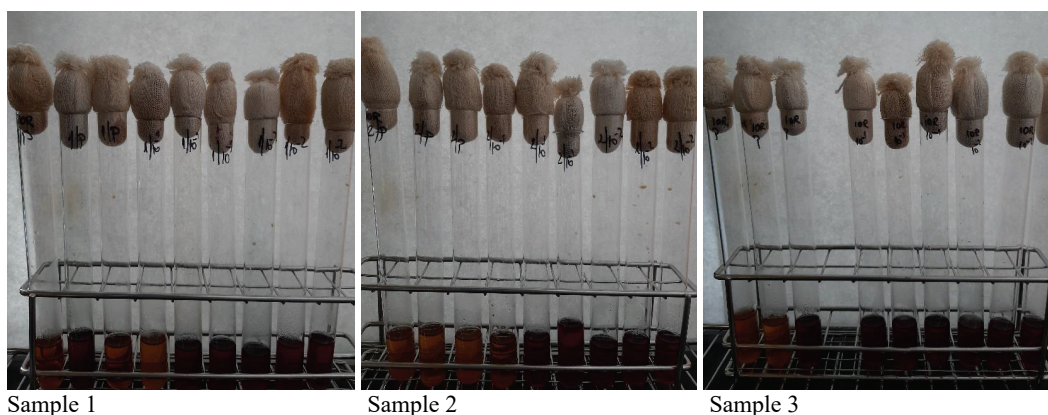


Figure 2. Highlighting the presence of faecal coliforms from water samples on the MacConkey culture medium.

From the group of microorganisms involved in the biogeochemical cycle of iron, the presence of ferrobacteria (which reduce  $Fe^{3+}$  to  $Fe^{2+}$ ) was highlighted in samples 1 ( $3.2 \times 10$  microorganisms/ml) and 2 ( $3.3 \times 10$  microorganisms/ml). Thus, colonies with red pigmentation were observed on the surface of the culture medium. The presence of ferrobacteria in water samples can be correlated with higher iron content, as it results from the XRF analysis of the samples. On the other hand, the UV-VIS spectra confirm the presence of carotenoids pigments which can argue for the brickish colour of lake water.

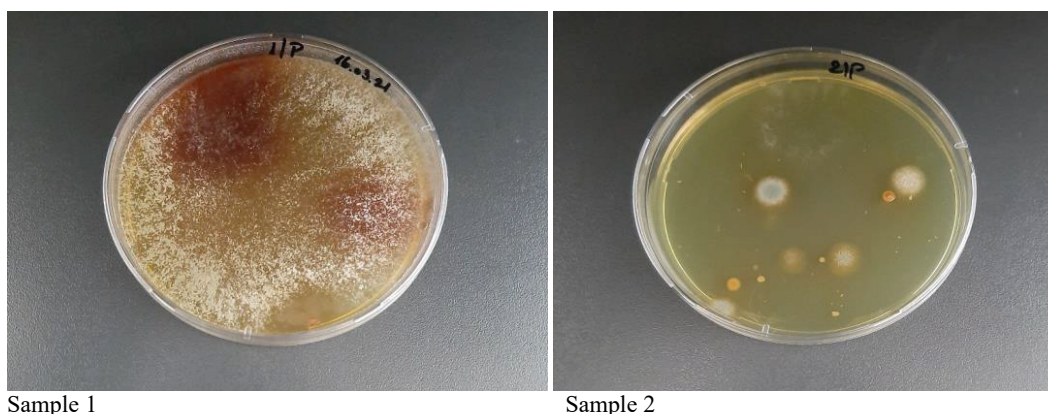


Figure 3. Evidence of the presence of ferrobacteria on Winogradsky culture medium.

### CONCLUSIONS

From the obtained data, the colour of the lake water is the result of its chemical composition, especially of a high concentration of iron compounds, as well as a result of the development of ferrobacteria that produce specific metabolites. The UV-VIS analysis revealed the presence of carotenoid compounds that also contribute to changing the colour of lake water throughout the mass.

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