Assessment of Nutrients-Chlorophyll-*a* Relationship in the Lower Danube River

Mihaela Ilie, Florica Marinescu, Gina Ghita, Ana-Maria Anghel, György Deák, Marius Raischi

***Abstract*—**Distribution and variability of chlorophyll-*a* were investigated in this study, in relation to nutrient concentrations (total nitrogen-TN, total phosphorus-TP) and hydro-meteorological conditions. The ratio and sum between TN and TP were also studied. Water samples were collected monthly, during September 2012 - August 2014, from left and right banks of the Danube River. Regarding the seasonal dynamics of Chl-*a*, the highest concentration was recorded in summer (49.75 µg/L) and the lowest one was observed in winter (0.59 µg/L). The highest Chl-*a* annual mean concentration was observed in 2013 (19.00 µg/L), closely linked to minimal discharge of the Danube River (2500 mc/s) from the monitored period. In case of Chl-*a* and total phosphorus**,** cluster analysis assembled the sampling sections in two different clusters considering the location. Pearson correlation coefficient revealed a weakly relationship between chlorophyll *a* and total nitrogen and no significant correlation between Chl-*a* and total phosphorus during summer.

***Keywords*—**chlorophyll *a*, cluster analysis, Danube River, nutrients

I. Introduction

Nutrients enrichment is a frequently mentioned cause for biotic deterioration of water bodies, but little is known about nutrients – chlorophyll-*a* (Chl-*a*) relationship in lotic environments. One of the most common ecological and environmental problems of water bodies is eutrophication, which diminishes water quality by spurring the excessive growth of algae and increasing suspended organic material [1, 2]. At present, there are more indicators of eutrophication, mainly being the nutrients as total phosphorus and total nitrogen and chlorophyll which is an indicator of phytoplankton biomass [3]. The growth of phytoplankton biomass in large rivers, mainly depends on hydrodynamic conditions, climatic conditions, residence time and light conditions but it is also influenced by the availability and concentration of nutrients [4, 5]. The levels of nutrients in a water body play an essential role in identifying the level of pollution [6]. The European Water Framework Directive 2000/60/EC (WFD), the most significant and complex legislative instrument in the field of water policy, develops the concept of ecological quality status for the assessment of water quality - based on the physical-chemical, hydro-morphologic and biological quality elements.

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Within the Danube River basin, phytoplankton assessment is particularly relevant because the Danube River as well as several of the larger tributaries have a great potential to produce large amounts of phytoplankton biomass [1]. In this study we evaluated the level of trophicity by means of the following parameters: total phosphorus, total nitrogen and chlorophyll-*a* and spatial representation of concentration variation for the investigated parameters was also performed.

II. Studied Area

This study investigates the relationships between Chl-*a*, total nitrogen (TN) and total phosphorus (TP) using data from a large range of samples from the Danube River between Km 347 and Km 182, during September 2012 - August 2014**.**

Average data on trophic state parameters, total phosphorus (TP), total nitrogen (TN) and chlorophyll-*a* (Chl-*a*) were obtained from the analysis of 560 water samples. Monthly samples were collected from ten sections (Fig. 1), between September 2012 and August 2014, from left and right banks of the Danube River.

The locations of sampling sections are shown in Table 1, and the sections were divided into two sectors: upstream (S1-S7) and downstream (S8-S10).

TABLE 1
Sampling Site Location Site Location

Sections	River km	Geographical coordinates (latitude, longitude)	
		Left bank	Right bank
S1	Danube km 348	44°10'35.63"N 27°32'18.44"E	44°10'18.31"N 27°32'33.18"E
S2	Bala Branch km 9.4	44°12'05.33"N 27°34'26.60"E	44°11'59.39"N 27°34'39.67"E
S3	Danube km 344.8	44°11'39.20"N 27°34'38.55"E	44°11'19.92"N 27°34'56.75"E
S4	Borcea Branch km 65	44°16'14.93"N 27°38'51.90"E	44°16'02.26"N 27°39'00.21"E
S5	Danube km 338	44°12'22.62"N 27°39'11.60"E	44°12'12.62"N 27°39'10.87"E
S6	Epurasu Branch km 1.8	44°11'25.64"N 27°41'08.09"E	44°11'18.12"N 27°41'10.72"E
S7	Danube km 334.3	44°11'53.25"N 27°42'10.45"E	44°11'37.96"N 27°42'11.23"E
S8	Caleia Branch km 8.9	45°04'56.17"N 27°54'06.61"E	45°04'53.16"N 27°54'21.88"E
S9	Danube km 186.5	45°08'39.15"N 27°57'43.51"E	45°08'43.19"N 27°57'52.69"E
S10	Danube km 182.6	45°10'19.60"N 27°56'22.34"E	45°10'21.23"N 27°56'46.71"E

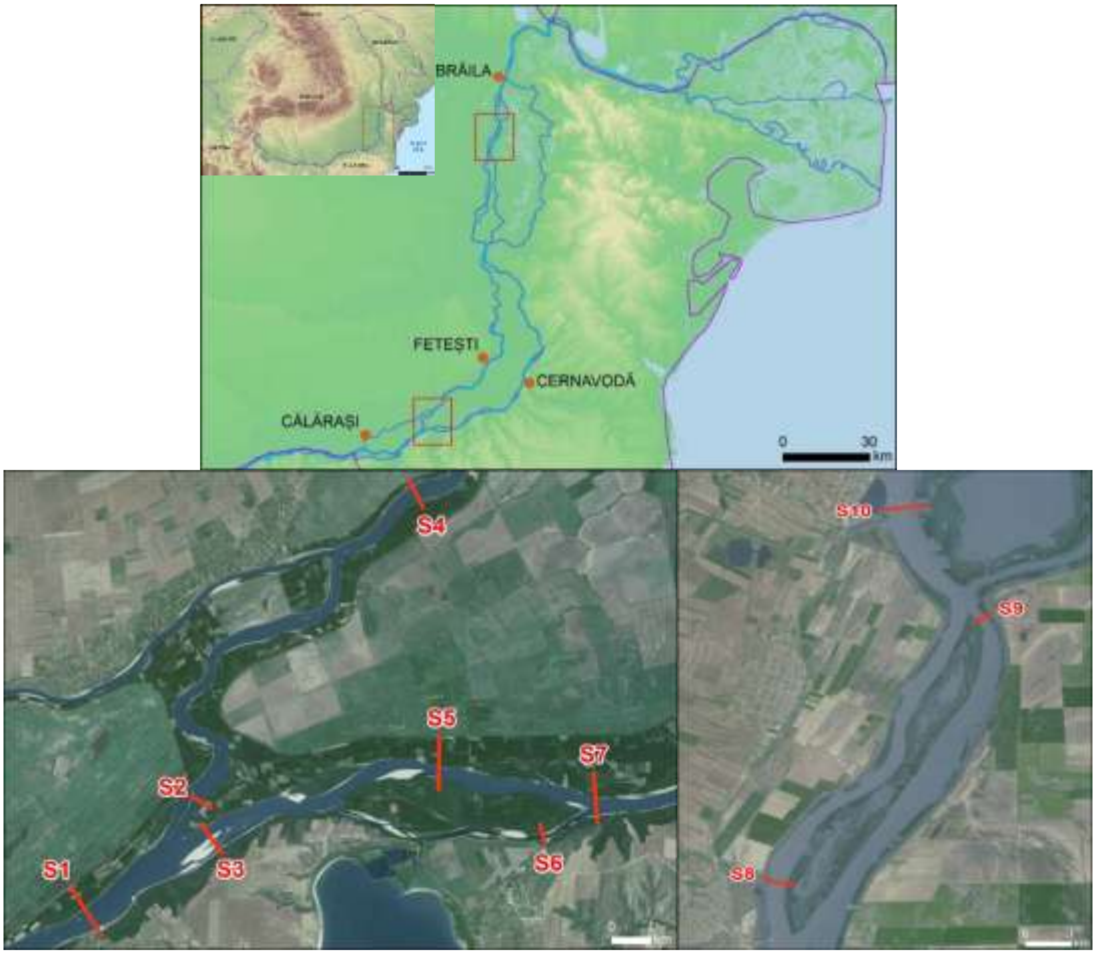

Fig. 1 Sampling sections located along the lower part of Danube River, Romania

III. Materials and Method

In order to assess the trophic status, water samples were taken monthly from ten sites along the Danube River Km 347 and Km Between 182 during September 2012 - August 2014 for analysis of NT, PT and chlorophyll-*a*. Sampling, processing and preservation of evidence WAS performed taking into account national and international Standards for determining TN, TP and chlorophyll-*a*. The river flow discharge (mc/s) of the Danube was measured in several points. Right after sampling, samples were placed in cold storage units, until further analysis in the laboratory.

Environmental and Water No 161/2006 for the Approval of the Norm Concerning the Reference Objectives for the Surface Water Quality Classification (Official Journal of Romania, Part 1, No 511 bis), transposed from European Water Framework Directive 2000/60/EC. The Order 161/2006 establishes five classes of environmental status for the aquatic ecosystems of lakes and rivers type: (I) very good, (II) good, (III) moderate, (IV) poor and (V) bad [7].

IV. Results and Discussion

Generally, nutrient concentrations were relatively high (1.64 mg/L average value of TN, 0.10 mg/L average value of

Chl-*a* was extracted using 90% of alcohol and spectrophotometrical quantification was performed at 665 and 750 nm. Nutrients (TN and TP) were also determined by spectrophotometrical method at 655 nm and 880 nm wavelengths, using the UV-VIS ATI UNICAM, type UV2 equipment.

Data for the studied parameters were analyzed using the descriptive statistics. Pearson correlation coefficient was used to correlate concentration values of TN, TP and Chl-*a* in the Danube. The cluster analysis was also used to distribute the studied parameters into groups based on similarities/ differences between different groups.

Water quality assessments were performed considering quality guidelines in Romania - Order of the Ministry of TP), with maximum values of TN and TP during summer (Fig. 2 and Fig.3). High values of chlorophyll-*a* concentration recorded in the summertime are associated with the low Danube flow (Fig. 4) as a result of low rainfall and relatively high concentrations of nutrients during this period. The averages of water temperatures was 15.0°C. The minimum water temperature was 1.1 °C in December and its maximum value reached 27.2°C in July.

The seasonal dynamics of Chl-*a*: the highest concentration was recorded in summer (49.75 μgL^{-1}) and the lowest one was observed in winter (0.59 μgL^{-1}).

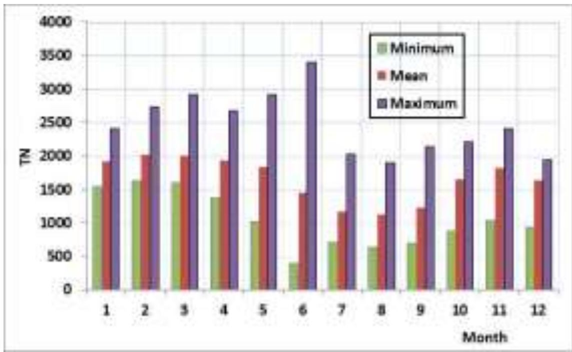

Fig. 2 TN concentration variation

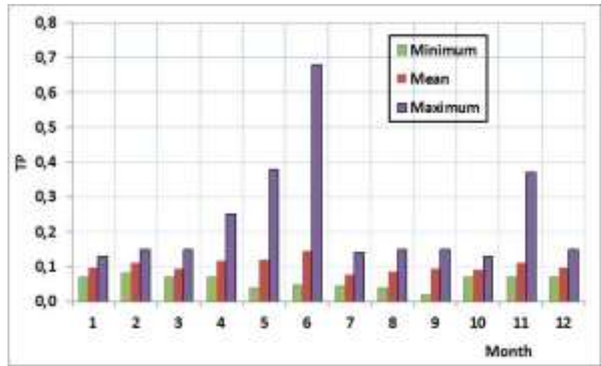

Fig. 3 TP concentration variation

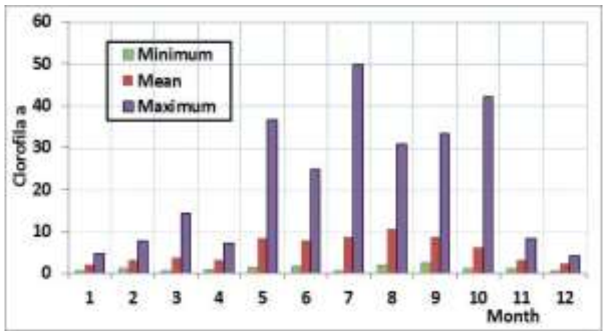

Fig. 4 Chl-*a* concentration variation

Statistical processing of analytical data and setting up the database to achieve GIS maps is an essential phase of water quality assessment. Statistical analyses were performed using the following software package: Minitab 16 and JMP 9 (SAS).

The ranges, means, standard deviations, and medians of TN, TP and chlorophyll-*a* during September 2012 - August 2014 are presented in Table 2.

In the area under study, the following concentration ranges were registered for the studied parameters: 0.390-3.390 mg / L for TN; 0.02-0.68 mg / L for TP; 0.59-49.75 mg / L for Chl-*a*. Similar range of nutrients and chlorophyll-*a* concentrations in water samples from the Danube was reported through the results obtained in the Joint Danube Survey Expedition 3 [8].

TABLE II
SUMMARY DESCRIPTIVE STATISTICS FOR TN, TP AND Chl-*a* DURING THE PERIOD 2012–2014

Variable	Season	Count	Mean	StDev	CoefVar	Median	Minimum	Maximum	CMA* Order 161/2006; WFD Limits [7,9]
TN	fall	80	1.7311	300.2	17.34	1.720	0.880	2.400	7.0
	spring	100	1.8688	329.6	17.64	1.840	1.009	2.910	
	summer	280	1.2478	473.3	37.93	1.130	0.390	3.390	
	winter	200	1.9114	282.4	14.77	1.900	0.930	2.910	
TP	fall	80	0.1014	35.21	34.73	0.095	0.070	0.370	0.4
	spring	100	0.1191	57.93	48.64	0.110	0.039	0.380	
	summer	280	0.1024	74.52	72.77	0.090	0.020	0.680	
	winter	200	0.0988	18.03	18.25	0.100	0.070	0.150	
Chl-a	fall	80	4,689	4,991	106,44	3,55	1,18	42,1	50
	spring	100	6,308	5,722	90,71	4,219	0,89	36,72	
	summer	280	8,889	5,869	66,03	7,7	0,59	49,75	
	winter	200	3,011	2,383	79,14	2,37	0,59	14,22	

Pearson correlation analysis was applied in order to correlate the studied parameters (tab. 3).

TABLE III
PEARSON CORRELATION (*r* VALUE) OF THE TN, TP AND Chl-*a* CONCENTRATION IN THE DANUBE SURFACE WATER

Season		TN	TP	Chl-a
2012-2014	TN	1		
	TP	0.372	1	
	Chl-a	-0.186	-0.02	1
spring	TN	1		
	TP	0.071	1	
	Chl-a	-0.184	0.355	1
summer	TN	1		
	TP	0.596	1	
	Chl-a	0.215	-0.128	1
fall	TN	1		
	TP	0.201	1	
	Chl-a	0.145	-0.09	1
winter	TN	1		
	TP	0.364	1	
	Chl-a	-0.048	-0.174	1

In the last decades, the clustering techniques have often been applied to a wide variety of research issues [10, 11, 12]. Cluster analysis: in case of Chl-*a* – TP the sampling sections were assembled in two different clusters considering the location.

Pearson's r coefficient and dendogram correlation for TN, TP and chlorophyll *a* concentration in the Danube water showed:

- a weak correlation between Chl-*a* and TP (r=0.355) in spring and TP-TN in fall (r=0.201) and winter (r=0.364);
- a moderate correlation between TP-TN (r=0.596) and a weak relationship between Chl-*a* and TP (r=0,215) during summer

The highest Chl-*a* annual mean concentration was observed in 2013 (19.00 µgL^{-1}), closely linked to minimal discharge of the Danube River (2500 mc/s) from the monitored period.

Pearson's r coefficient revealed a weak relationship between TP and TN (r=0.372) and no correlation between Chl-*a* - TN and Chl-*a* - TP for the entire monitoring period.

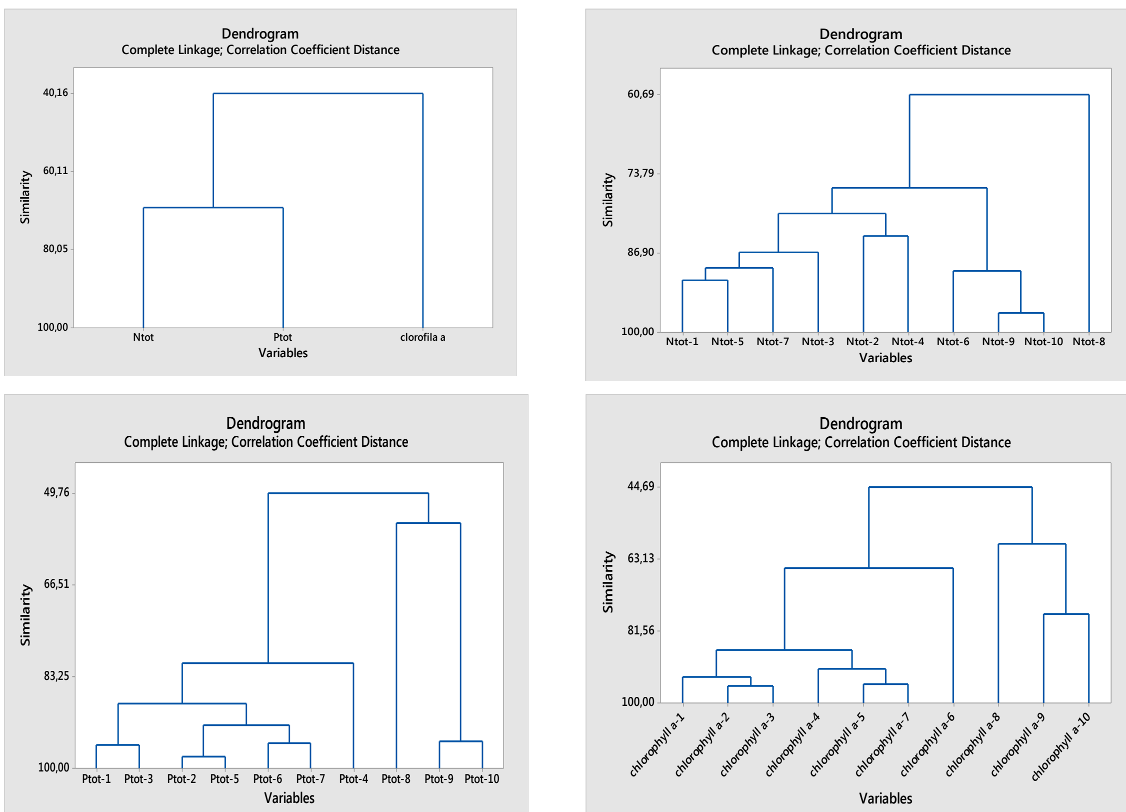

Fig. 5 Dendrograms of the water sampling sections from the Danube River

Spatial distribution of NT, TP and Chl-a in water is important to assess the variation of nutrient quantities due to the influence of human activities [13], hydrological conditions, morphodynamics [14] and nutrient retention.

The GIS mapping technique was involved to create spatial distribution maps of TN, TP and Chl-*a* in water samples from the Danube River as well as the Danube flow variation during September 2012 - August 2014. The software used for mapping and spatial analysis was ArcView 9.3. These operations consist of interpolation of all values and developing spatial images (map) to highlight the values (value classes) by cartographic ways. Spatial distribution of TN, TP and Chl-*a* in water along the Danube River between Km 347 and Km 182 as well as the Danube flow variation are shown in the following figures.

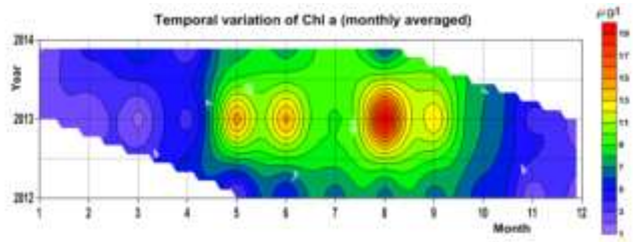

Fig. 6 Temporal variation of Chl-a in the Lower Danube River (monthly averaged in September 2012 - August 2014)

Distribution of Chl-*a* shows high temporal and spatial variations (Fig 6), and the main factors controlling its distribution during summer, were TN and TP (Fig.7) availability and the low Danube flow due to low rainfall in summer. High values of chlorophyll concentration recorded in the summertime are associated with the low Danube flow (Fig.8) as a result of low rainfall and relatively high concentrations of nutrients during this period.

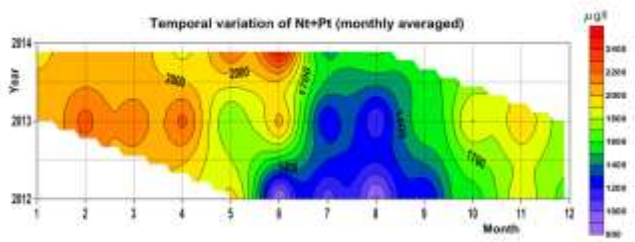

Fig. 7 Temporal variation of the TN+TP in the Lower Danube River (monthly averaged during September 2012 - August 2014)

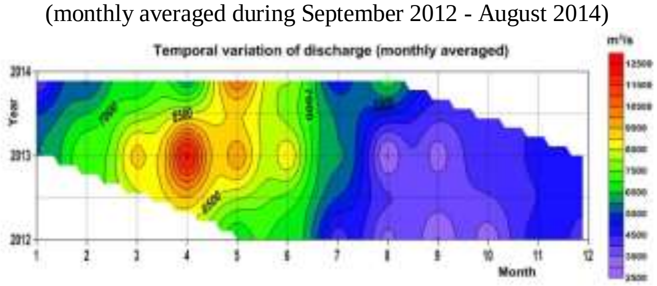

Fig. 8 Temporal variation of the Danube River discharge (monthly averaged during September 2012 - August 2014)

V. Conclusion

Eutrophication of surface waters is a consequence of enriching water with nutrients and leads to overgrowth of algae and other aquatic plants. This study was conducted to assess changes in concentrations of nutrients and Chl-*a* by determining their concentration and spatial distribution in the Danube River, between Km 347 and Km 182, during September 2012 - August 2014. It reveals that in terms of Chl-*a*, there are significant seasonal variations, with maximum concentrations reached in the summertime, when biological activity is most intense, being strongly influenced by weather conditions and anthropogenic pressures.

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