

PHOSPHATASE ACTIVITY OF WATER AS A PARAMETER OF THE RIVER TISA WATER MONITORING

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Abstract

In this paper the results of microbiological and enzymological examination of the Yugoslav part of the river Tisa water are presented. During the period from 1983 to 1986 the water of five localities was analyzed.

Classical microbiological parameters, such as total number of planktonic bacteria, number of heterotrophic bacteria, and T/H index of saprobity (the ratio between the total number of planktonic bacteria and the number of heterotrophic bacteria), being conventionally used as indicators of the level of saprobity of surface freshwaters, were compared with the index of phosphatase activity, a new indicator of the water eutrophication degree.

Statistically significant correlation between classical microbiological and the new biochemical indicator of organic load of the river Tisa affirms the index of phosphatase activity as a reliable parameter for a fast preliminary estimation of the saprobity degree of surface freshwaters.

Introduction

In recent years the investigation of metabolic activity has been increasingly used in assessment of both trophic level and the degree of pollution of certain aquatic ecosystems. JONES (1972) proposed measurement of relative phosphatase activity as an indicator of biomass and general trophic conditions of aquatic habitats suggesting that water enzymatic activity may serve as a useful indicator of surface water eutrophication.

Most microorganisms synthesize phosphatases, enzymes that catalyze orthophosphate release from organic phosphomonoesters. This was confirmed by our previous studies (MATAVULJ *et al.*, 1976; 1978).

In the microbiological study of eutrophic marine water of Tokyo Bay, TAGA and COBORI (1978) found positive correlation between the activity of alkaline phosphatase and standard indicators of water quality (the number of heterotrophs, count of phosphatase producers, inorganic and organic phosphate content, the amount of proteins, chlorophyll and DNA).

JONES (1972) gave the equation which involves phosphatase activity with total phosphate content and total biomass. Various biochemical parameters, particularly the activities of some enzymes (phosphatase, proteinase, cellulase) are already being used in estimate of trophic level and degree of pollution of surface waters (VERSTRAETE *et al.*, 1976; KARPUSHIN and MELJNIKOV, 1972; ZDANOVSKI, 1977).

Our previous studies on phosphatase activity of waters with different degrees of eutrophication suggested possible use of water enzymatic activity as an indicator of its organic load (MATAVULJ *et al.* 1983; 1984a; MATAVULJ and FLINT, 1987). In this paper, the river Tisa with characteristic changes of water saprobity degrees (depending on locality and season) served as an object for check and affirmation of phosphatase activity index, the new biochemical parameter used in water condition estimates.

Materials and Methods

Microbiological assays comprised standard, cultivating, and direct methods. The total count of planktonic bacteria was determined by ultrafiltration on Sartorius membrane filters with pore diameters of 0.2 μm , after RAZUMOV (1932). The colonies of aerobic heterotrophic bacteria were counted on plates of Nutrient Agar (Torlak MPA).

Phosphatase activity was measured with p-Nitrophenyl-phosphate (pNPP) as a substrate by the modified method, described by FLINT and HOPTON (1977). Enzymological analyses preceding calculation of water phosphatase activity index (MATAVULJ, 1986) included measurement of total activity of acid, neutral and alkaline phosphatases in an original (untreated) water sample.

Results and Discussion

The counts of saprophytic bacteria, represented as means of the results from four years of study, show that water of the river Tisa belonged to 2nd—3rd class after Kohl's (1975) categorization (it belonged to 3rd class only at Titel). From Fig. 1 it may also be seen that, as a rule, water quality is better in early summer than in autumn.

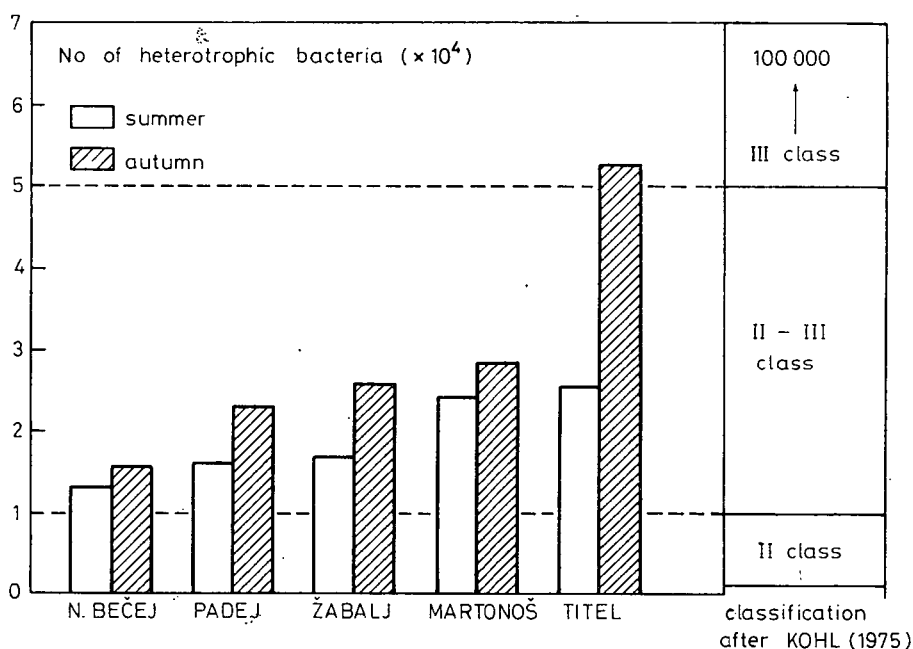


Fig.1

Judging by T/H index of saprobity (the ratio between total number of planktonic bacteria and number of heterotrophic bacteria) the same conclusion may be drawn, i.e. water from all sectors tested was of better quality in early summer than in autumn. According to this criterion, water of the river Tisa belonged to moderately polluted and/or polluted waters.

Our long-term comparative investigations of standard microbiological and biochemical indicators of water quality, with respect to its organic load, and new enzymological indicators of saprobity degree e.g. the level of eutrophication of different surface waters (MATAVULJ *et al.* 1982; 1984b; MATAVULJ, 1986) resulted in the proposed introduction of a new system of categorization of surface freshwaters, regarding degrees of their organic load, on the basis of the index of phosphatase activity of water (PAI). PAI represents a mean value of the acid, neutral, and alkaline phosphatase activities of original water sample (Table 1).

Tab. 1. *The proposal of a system of surface freshwater categorization regarding degrees of their organic matter load, based on the water enzymic activity, i.e. index of water phosphatase activity (MATAVULJ, 1986)*

Phosphatase activity index ($\mu\text{mol/s/dm}^3$ pNP) 30 °C	Proposal of category name		Characteristic of water (conditionally)	Equivalent to classes according to		
				Kohl (1975)	Tüm-kling (1969)	Pantle—Buck (1955)
below 0.01	I	A	MAXIMALLY CLEAN	I	I	KATAROBIC ZONE
0.01—0.10		B	VERY CLEAN			
0.10—0.25	I—II		CLEAN			
0.25—0.50	II	A	SATISFACTO-RILY CLEAN	I—II	II	ÖLIGO-SAPROBIC ZONE
0.50—1.00		B	SLIGHTLY POLLUTED	II		
1.00—2.50	II—III		MODERATELY POLLUTED	II—III	III	β -MESO SAPR. ZONE
2.50—5.00	III	A	POLLUTED	III		β - α MESO-SAPROBIC ZONE
5.00—7.50		B	VERY POLLUTED	III—IV		
7.50—10.00	III—IV		DIRTY	IV	IV	α -MESO-SAPR. ZONE
10.00—15.00	IV	A	VERY DIRTY			
above 15.00		B	MAXIMALLY DIRTY			

Statistically significant correlation between heterotrophic count as a standard indicator of water quality and a new enzymological parameter may be observed from the Fig. 2. Somewhat lower degree of correlation was recorded between PAI and the total number of bacteria (Fig. 3) which is probably due to the fact that milipore-filter bacterial count includes dead, dormant, and inactive cells. This is consistent with the results of our previous investigations of natural surface waters (MATAVULJ *et al.* 1982; 1984a; MATAVULJ, 1986).

PHOSPHATASE ACTIVITY INDEX (pNP $\mu\text{mol/s/dm}^3$)

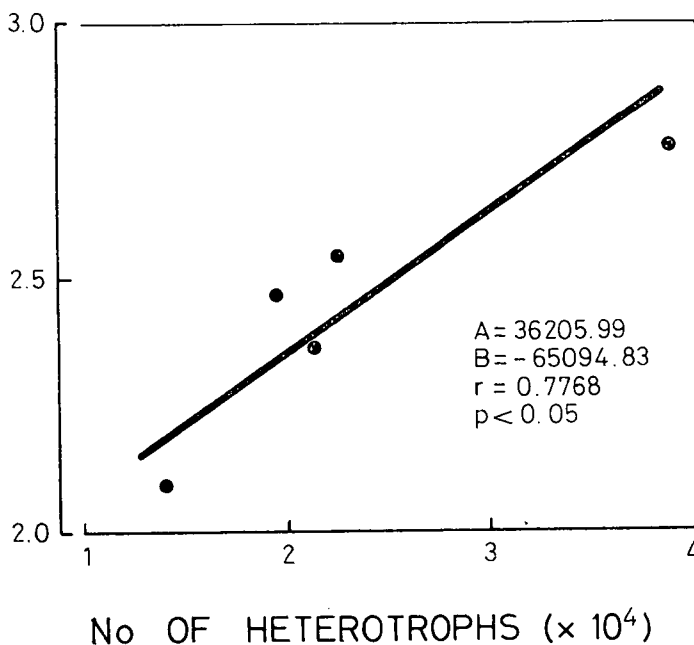
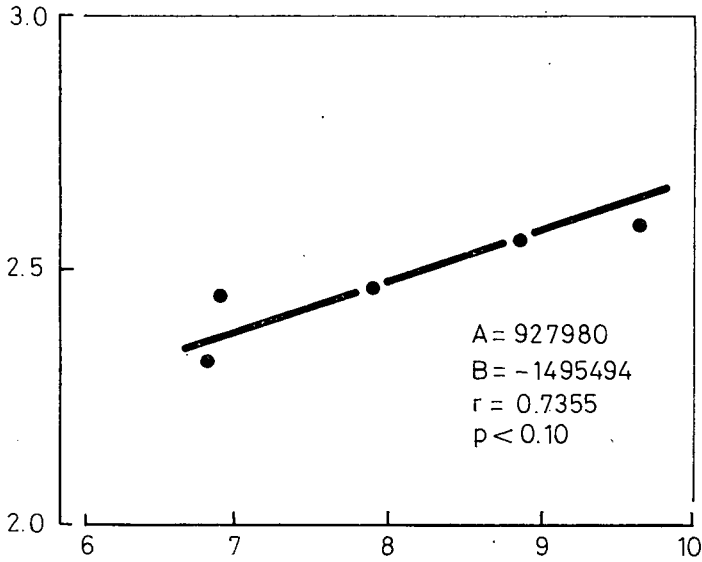


Fig. 2. Correlation between the viable bacteria count and water phosphatase activity index

Fig. 4 illustrates the negative correlation between PAI and T/H index of saprobity. This phenomenon points out positive correlation between the level of enzyme activity and the water saprobity degree, the latter being represented as T/H index. Statistically significant degree of correlation between neutral phosphatase activity and T/H index of saprobity (Fig. 5) deserves to be specially emphasized. Consequently, when calculating PAI as an indicator of organic pollution of surface freshwaters, neutral phosphatase activity should be taken into account.

Considering that phosphatases are primarily indicators of water pollution with organic phosphates, our proposed classification of surface freshwaters according to degree of organic load on the basis of PAI is only conditional. We deem that an index of water enzymic activity would be a much more reliable indicator of water organic pollution if it also comprised the levels of activities of some other enzymes such as proteinases, dehydrogenase, lipases, saccharases, cellulases, etc. In this respect, we have started examining β -glucosidase activity of the river Tisa water, since

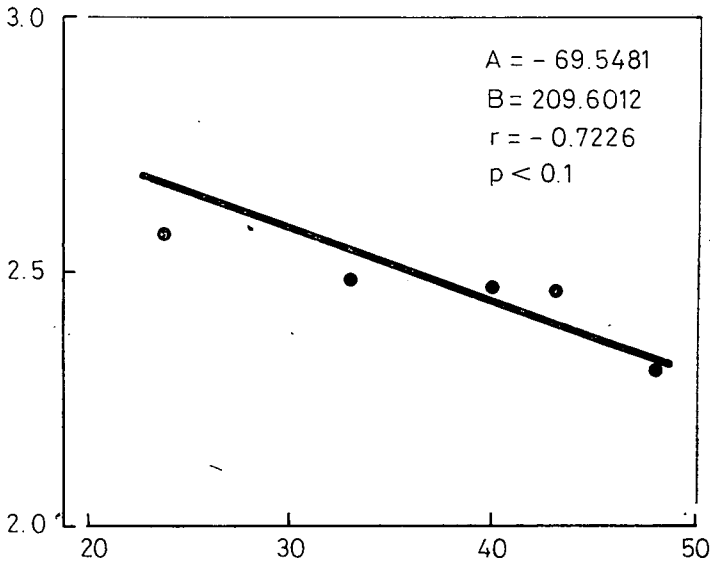
PHOSPHATASE ACTIVITY INDEX (pNP $\mu\text{mol/s/dm}^3$)



TOTAL No OF PLANKTONIC BACTERIA ($\times 10^5$)

Fig. 3. Correlation between the total number of bacteria and water phosphatase activity index

PHOSPHATASE ACTIVITY INDEX (pNP $\mu\text{mol/s/dm}^3$)



T/H INDEX OF SAPROBITY

Fig. 4. Correlation between the T/H index of saprobity and water phosphatase activity index

NEUTRAL PHOSPHATASE ACTIVITY (pNP $\mu\text{mol/s/dm}^3$)

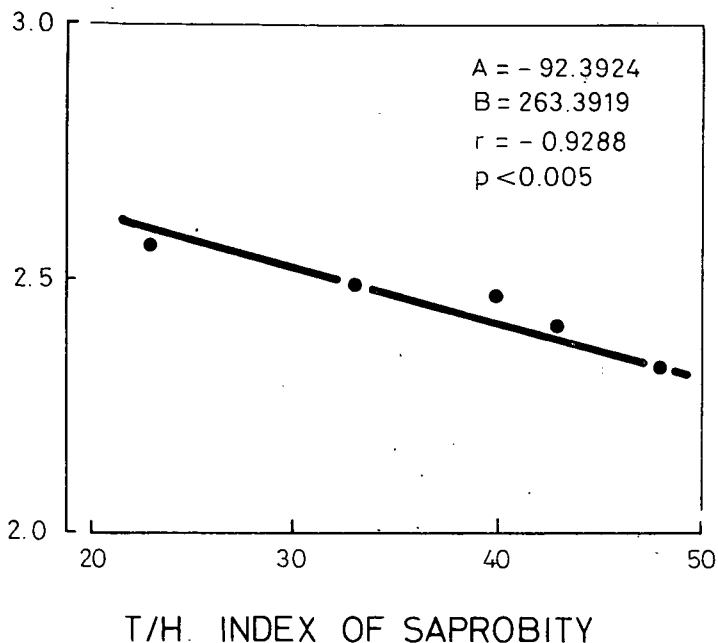


Fig. 5. Correlation between the T/H index of saprobity and water neutral phosphatase activity

most aerobic saprophytes have this enzyme which is involved in polysaccharide biodegradation. The results of these investigations were presented at the 17th scientific meeting on the river Tisa (MATAVULJ *et al.* 1986) and will be published in the following paper.

Conclusion

Statistically significant correlation between standard microbiological indicators of water saprobity and new, enzymological indicators of water eutrophication degree points out possible use of phosphatase activity of water as a biochemical parameter which might enable faster and more reliable assessment of organic load of surface freshwaters.

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A víz foszfátáz aktivitása mint a Tisza vízének monitoring paramétere

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Kivonat

A szerzők a Tisza jugoszláv részlegének martonos—padé—töröbce—zsablya—titeli szakaszán 1983—1986-os időszakban végzett mikrobiológiai és enzimológiai vizsgálatának eredményeit ismertetik. A felszíni vizek szaprobitásának meghatározása hagyományos mikrobiológiai paraméterek (a baktériumplankton teljes száma, a heterotrof baktériumok száma, a T/H szaprobitási index) és a víz foszfátáz aktivitási indexének mint újkeletű szaprobitási indikátornak összevetésével történt. A Tisza vízének szerves anyagokkal való megterhelése az összevetett paraméterek (a hagyományos és új biokémiai indikátor) által kapott, statisztikailag szignifikáns korrelációs mutatója arra utal, hogy a víz foszfátáz aktivitási indexe mint megbízható paraméter elősegítené a felszíni vizek szaprobitási fokozatának gyorsabb és megbízhatóbb meghatározását.

Фосфатазная активность воды как параметр мониторинга воды реки Тиса

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Резюме

В статье представлены результаты микробиологических и энзимологических исследований, выполненных на реке Тиса в югославской части. В периоде с 1983 по 1986 г.г. анализу подвергались пробы воды, отбор которых осуществлялся в пяти пунктах (места: Мартонош, Падей, Нови Белей, Жабаль и Тител).

Классические микробиологические параметры, в том числе общая численность бактериопланктонов, количество гетеротрофных бактерий и Т/Х индекс сапробности (отношение между общим количеством планктонных бактерий и количеством гетеротрофных бактерий, установленными на питательном агаре), которые стандартно применяются как индикаторы уровня сапробности поверхностных пресных вод, сравнивались с индексом фосфатазной активности воды, новым индикатором степени сапробности воды.

Статистический показатель соотношения между классическими микробиологическими и новыми биохимическими индикаторами органической нагрузки воды р. Тиса, подтверждает индекс фосфатазной активности воды как надежный параметр более быстрой и достоверной предварительной оценки степени сапробности поверхностных пресных вод.

Fosfatazna aktivnost vode kao parametar monitoringa vode reke Tise

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Rezime

U radu su prikazani rezultati mikrobioloških i enzimoloških istraživanja jugoslovenskog dela reke Tise. U periodu od 1983 do 1986. godine analizirani su uzorci vode sa pet lokaliteta (Martonoš, Padej, Novi Bečej, Žabalj i Titel).

Klasični mikrobiološki parametri kao što je ukupna brojnost bakterioplanktona, broj heterotrofних bakterija i T/H indeks saпрobности (odnos između ukupnog broja planktonskih bakterija i broja heterotrofних bakterija utvrđenog na hranljivom agaru), koji se standardno koriste kao indikatori nivoa saпрobности površinskiх slatkih voda, poredjeni su sa indeksom fosfatazne aktivnosti vode, novim indikatorom stepena saпрobности vode.

Statistički sginifikantna korelacija između klasičnih mikrobioloških, i novih biohemijskih indikatora organskog opterećenja vode reke Tise, afirmiše indeks fosfatazne aktivnosti vode kao pouzdan parametar za bržu i verodostojniju preliminarnu procenu stepena saпрobности slatkih površinskiх voda.