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The activity of bacterial flora in the River Nida, in stretches differing in geological substratum and soils near the banks (Southern Poland)*

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Abstract — The production of bacterial biomass and the magnitude of its bioaccumulation and biodegradation were determined for stretches of the river differing in the kind of soil near the banks. It was found that the amount of energy used for bioaccumulation and biodegradation increased in the lower part of the catchment area of the Nida. It was demonstrated that the trophic level of the water increased with the course of the river, this being indicated by a fall in the value of Schrödinger's coefficient and a rise in the coefficient of heterotrophic activity, determined in relation to asparagine.

Key words: river, bacteria, activity, biomass production, bioaccumulation, respiration.

1. Introduction

The microbial activity in surface waters is a mesure of the magnitude of the energy flow and of the cycling of organic matter in these environments; it reflects the biodegradation of organic matter, and the cycling of nutrients in a given biological system (Oppenheimer 1960, Jewell et al. 1971). It also points to the relationship between bacterial

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assimilation and the nutrient concentration in the water (Wright, Hobbie 1965, 1966, Vaccaro, Janash 1966, Wetzel 1967, Trzilova, Miclošovičova 1979).

Organic matter dissolved in water is taken up mainly in bacterial processes (Sepers 1977, Kang, Seki 1983). Therefore, determination of the bioaccumulation and respiration at the level of heterotrophic bacteria, which are a major dominant among aquatic organisms, reflects the direction taken by processes connected with the synthesis or decomposition of organic material. Hence, it also indicates the trophic level, or the degree of water pollution.

The aim of the present study was to determine the activity of aquatic bacteria, under the influence of substances flowing down from the surrounding land, on stretches of rivers differing in the type of soils near the bank..

It is known that the run-off into a river of substances from the surrounding area which have either come into being naturally in the drainage basin or which have been introduced into it artificially (e.g. by various agricultural measures or sewage disposal) is determined to a considerable degree by the kind of soils close to the banks (Margowski 1976).

2. Study area

The River Nida, which belongs to the catchment area of the Vistula, drains a depression (called the Nidziańska syncline) between the Silesian-Krakôw Upland and the Kielce Upland. It issues from the confluence of the Rivers Czarna Nida and Biała Nida and is characterized by a varying degree of water purity (Pasternak 1973, Pasternak, Starzecka 1979, Starzecka 1979, Starzecka et al. 1979), and differing habitat conditions on the particular stretches. It has been found that these conditions affect the formation of associations of sessile algae, and also the micro- and macrofauna (Dumnicka 1978, Srokosz 1980, Grabacka unpubl. data, Kawecka 1986).

The investigations were carried out in the upper and lower drainage basin of the Nida, at four stations: on the Czarna Nida at Morawica (Station 1), on the Biała Nida at Mniszek (Station 2), and on the Nida at Kowala (Station 3) and Chroberz (Station 4) (fig. 1). Table I gives the type and the textural group of the soils near the banks, described on the basis of data obtained from the Department of Microbiology of the Agricultural Academy in Kraków.

The substratum of the Czarna Nida is composed mainly of dolomites, limestones, and sandstones of the Devonian and Jurassic, and that of the Biała Nida and Nida of marls and limestones (Pasternak 1973).

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Fig. 1. Catchment area of the River Nida. 1—4 — stations

Table I. Description of soils near the banks in the catchment area of the Nida

No of station	River	Locality	Soil: type and subtype	Soil: family	Soil: soil textural group
1	Czarna Nida	Morawica	brown, medium deep rendzina	formed from mid-Triassic limestone	medium deep sandy loam, on loamy limestone weathering waste
2	Biała Nida	Mniszek	peaty alluvial soil	formed from peaty river alluvia	medium deep, low peat, on sand
3	Nida	Kowala	medium deep, humic rendzina	formed of Upper-Cretaceous marl	deep, heavy loam on loamy, skeletal weathered waste
4	Nida	Chroberz	typic (humic), heavy alluvial soil, pseudogleyed and gleyed	formed from alluvia of the River Nida	very fine sandy soil and medium-very-fine-sandy loam on heavy and medium. very fine sandy, medium deep loam, on sand interbedded with silt and sand

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The upper stretch of the Czarna Nida is characterized by pure water with a large quantity of dissolved oxygen, low oxidability, a low chloride, potassium, and sodium content and also a low content of ammonia, nitrites, and phosphates (Pasternak 1973, Pasternak, Starzecka 1979, Starzecka et al. 1979).

The water of the Biała Nida is well oxygenated, with a low potassium, sodium, magnesium, chloride, and phosphate content, a medium iron content, and a calcium content considerably higher than that in the Czarna Nida. This can be attributed to the differences in the geological structure of the substratum (dominance of chalk and Jurassic limestone). A higher oxidability, higher BOD_5 values, and a more intense colour than in the pure waters of the Czarna Nida are also typical of it. An increase in the indices reflecting the content of organic matter and the intensity of the colour of the water is associated mainly with the presence of humic compounds in the water, which (especially in the spring period) are washed out in large quantities from the peat soils which cover large expanses of the catchment area of the Biała Nida (Pasternak 1973, Pasternak, Starzecka 1979).

The water of the lower reaches of the Nida shows a lower content of dissolved oxygen, high oxidability, higher BOD_5 values, and a large content of calcium, potassium, magnesium, sulphates, chlorides, and nutrients. As compared with the Czarna Nida, the water of the Nida has a higher content of trace elements, such as Zn, Cr, Mn, Pb, Co, and Cd (Pasternak 1973).

From the results obtained during the study, it was found that the water at all the stations was of calcium-hydrocarbonate type, with a distinctly higher content of anions and cations (by 65%) in the lower part of the catchment area (Bombówna unpubl. data).

3. Material and methods

The study concerned mixed associations of planktonic bacteria of the Rivers Czarna Nida, Biała Nida, and Nida in samples of water taken on 25 dates, at monthly intervals, in the period from February 1982 to February 1984.

The activity of the bacterial microflora was determined on the basis of the depletion of oxygen in cultures kept in natural (not enriched) water, and in cultures with added asparagine (100 μM of asparagine per dm^3 of water). In order to remove phyto- and zooplankton, the water was filtered through a No 25 plankton net, then poured into 250 ml flasks with ground-in stopperes and incubated at a temperature of approximately 22°C. After 24 hours, the oxygen content in the cultures was determined using a GXI 610E oxygen electrode. The production of bacterial biomass

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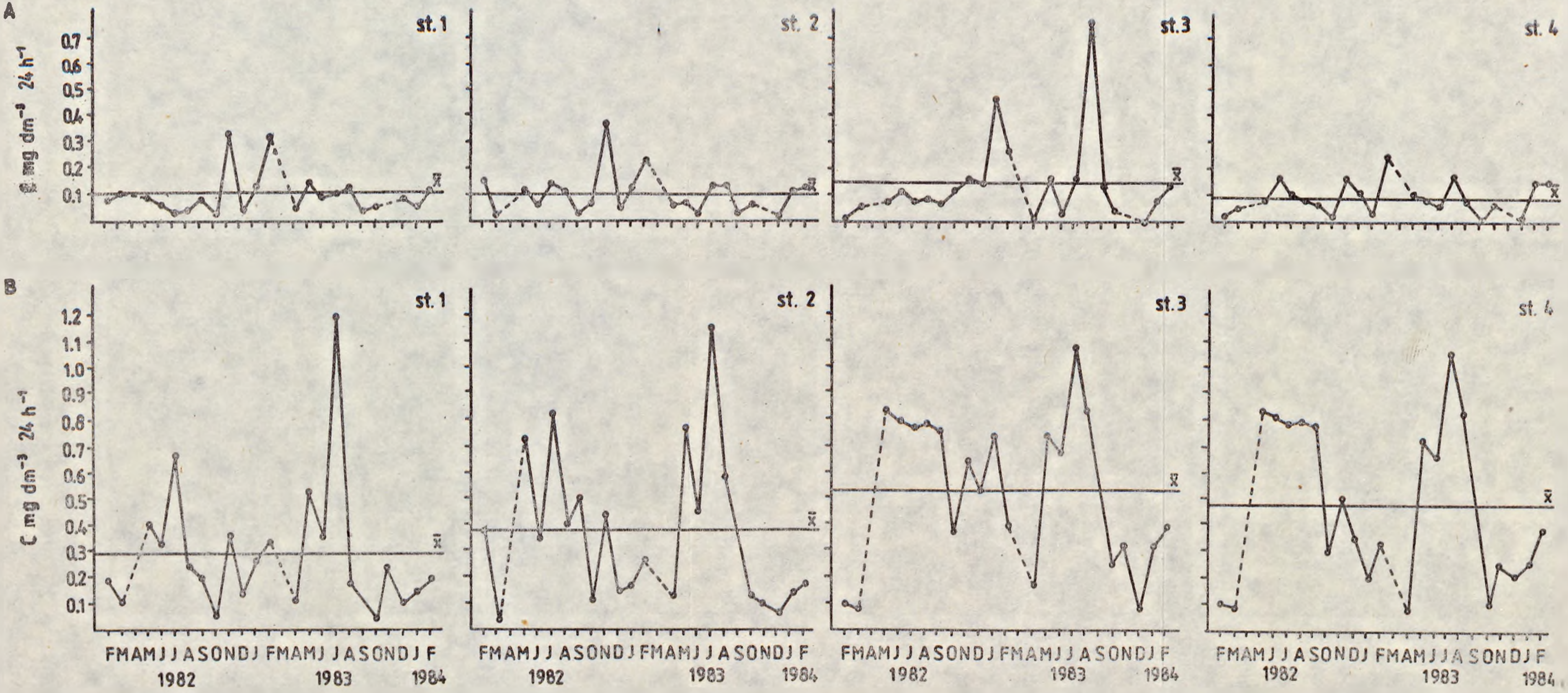

Fig. 2. Seasonal changes in the production of bacterial biomass in the period 1982—1984 at stations 1—4. A — natural water without substrate; B — water high added asparagine (100 μM dm^{-3}); — mean of the investigation period; 1—4 — stations

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(in C mg dm^{-3} water 24 h^{-1}) was calculated from the amount of oxygen taken up (Sorokin, Kadota 1972).

The bacterial biomass, in mg dry weight per dm^3 of water, was calculated according to Kuznyetsov and Romanyenko (1963). The number of bacteria per ml of water (means from the study period), determined by the plate method, was used for the calculations. A correction was made to allow for the fact that the results obtained by this method were understimated, by multiplying the number of bacteria determined by a factor of 10 (Starzecka 1988, in press).

Schrödinger's coefficient, expressing the ratio of respiration to biomass, which describes the energy expenditure per unit biomass (Odum 1982), was calculated. For this purpose, the respiration and biomass values obtained were converted into calories, assuming that 1 mg O_2 is equivalent to 3.38 calories and 1 mg dry weight to 5 calories.

The coefficients of heterotrophic activity were calculated from the ratio of production of bacterial biomass in cultures with asparagine to that of biomass in control cultures kept in natural (not enriched) water.

4. Results

4.1. Production of bacterial biomass

The production of bacterial biomass was found to be most constant in the natural water of the Nida at Station 4 (0.01—0.25 C mg dm^{-3} 24 h^{-1}), fluctuations being greatest at Station 3 (0.01—0.76 C mg dm^{-3} 24 h^{-1}), while in the Czarna Nida (Station 1) and the Biała Nida (Station 2) the fluctuations were smaller, amounting to 0.02—0.34 and 0.02—0.36 C mg dm^{-3} 24 h^{-1}, respectively (fig. 2A).

The mean production in the study period, in the order of 0.1 C mg dm^{-3} 24 h^{-1} was similar at Station 1, 2, and 4, but at Station 3 it was higher (50% on average) (fig. 2A).

An increased activity of the bacterial microflora of the water of the Czarna Nida (Station 1) and Biała Nida (Station 2) was recorded in November 1982 and in February 1983. For the water of the Nida (Stations 3 and 4), this was observed in January and February 1983 and also in August of the same year, at Station 3. On the remaining sampling dates, biomass production was close to or lower than the mean from the whole study period (fig. 2A).

From the results obtained for particular seasons, it can be seen that the highest biomass production occurred in summer at Stations 2, 3, and 4 and in winter at all stations. In the spring, the activity of the bacterial microflora was several times smaller (Table II).

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Table II. Production of bacteria biomass C mg dm^{-3} 24 h^{-1} (mean of seasons). K - natural water (without substrate); A - water with added asparagine (100 µM dm^{-3})

No of Station	Spring		Summer		Autumn		Winter	
	K	A	K	A	K	A	K	A
1	0.096	0.298	0.079	0.553	0.096	0.176	0.121	0.203
2	0.060	0.418	0.122	0.699	0.102	0.356	0.115	0.216
3	0.072	0.495	0.277	0.871	0.108	0.439	0.179	0.361
4	0.075	0.481	0.098	0.877	0.064	0.354	0.083	0.236

In the cultures kept in water with asparagine, bacterial biomass production varied from 0.04—1.20 C mg dm^{-3} 24 h^{-1}. The greatest range of variation was found at Station 1 (1.15 C mg dm^{-3} 24 h^{-1}), a slightly smaller one at Station 2 (1.12 C mg dm^{-3} 24 h^{-1}), and at Station 3 (1.00 C mg dm^{-3} 24 h^{-1}) and 4 (0.99 C mg dm^{-3} 24 h^{-1}) (fig. 2B).

In cultures with asparagine, bacterial activity was highest in summer, a marked increase in biomass production being recorded in July and August 1983. Increased biomass production was also typical in spring (fig. 2B, Table II).

On the basis of the mean production of biomass in cultures with asparagine, calculated for the whole 1982—1984 study period, the stations may be divided into two groups — the first with Stations 1, 2 and a biomass production of 0.29 and 0.37 C mg dm^{-3} 24 h^{-1}, respectively, and the second with Stations 3, 4 and a biomass production of 0.53 and 0.48 C mg dm^{3-} 24 h^{-1} (fig. 2B).

4.2. The energy flow at the level of associations of heterotrophic bacteria

In the water studied, the flow of energy was analysed on the basis of the amount of energy accumulated in the biomass of heterotrophic bacteria and the amount of energy released from the environment as a result of biooxidation of organic substances.

In the water of the Czarna (Station 1) and Biała Nida (Station 2), the amounts of energy accumulated in bacterial cells were similar and on average about 4 times smaller than the values obtained from the water of the Nida (Stations 3 and 4).

Bacterial respiration, including the amount of energy released from the environment, was highest at Station 3, being lower in the two upper Stations (1 and 2) and lowest at Station 4 (fig. 3).

The values of S c h r o d i n g e r 's coefficient, indicating the expenditure of energy per unit biomass, were similar for the waters of the upper reaches of the Nida (Stations 1 and 2) and distinctly higher than those recorded from Stations 3 and 4 in the lower reaches of this river (fig. 3).

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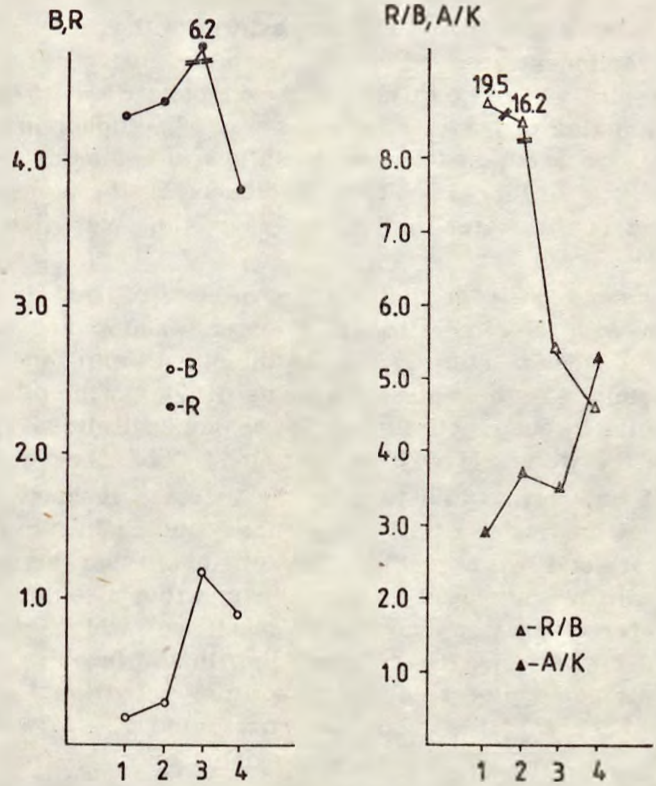

Fig. 3. Mean biomass content, respiration rate of bacteria, and coefficients R/B, A/K in the period 1982—1984, at particular stations (1—4). B — biomass (cal dm^{-3}); R — respiration (cal dm^{-3} 24 h^{-1}). Production of bacterial biomass: A — in the water with asparagine (100 μM dm^{-3}); K — (control) in the natural water, without substrate

The coefficient of heterotrophic activity, describing the potential activity of the bacterial microflora, was much the same for the waters of the Czarna Nida, (Station 1) Biała Nida (Station 2) and Nida at Station 3, and slightly higher for the water of the Nida at Station 4 (fig. 3).

5. Discussion

The results obtained in the present study show that the production of biomass in the waters of the Czarna Nida (Station 1), Biała Nida (Station 2), and Nida (Station 4) is very similar. Despite some seasonal differences, the mean production from the study period was fairly even, amounting to 0.1 C mg dm^{-3} 24 h^{-1}. On the one hand, this indicates

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a greater abundance of nutrient compounds in the lower part of the river and on the other a greater activity of bacteria in the water of the upper part of the catchment area, of which a smaller supply of nutrient compounds is typical (Pasternak 1973, Starzecka 1979). The fact that the production of bacterial biomass was 50% higher in the Nida at Station 3 may be accounted for by the inflow of communal wastes from the up-river town of Pińczów. This is evidenced by the higher phosphate concentration in the water in this section of the river (Bombówna unpubl. data).

It is common knowledge that the biosynthesis of bacteria takes place concurrently with the mineralization of organic matter and is a function of the bacterial assimilation of mainly dissolved, auto- and allochtonic material contained in the water (Sepers 1977). On the other hand, the bacteria exhibit greater activity at lower concentrations of substrate available in the water (Hobbie, Wright 1965, Sepers 1977).

It would be worth while to note the increase in bacterial biomass production in the water of all the stations studied in November 1982, and in January and February 1983. It is very likely that this increase was connected with the increased surface flow and thus also with the release into the water of greater amounts of substances which may be easily decomposed by bacteria. It was found that the surface run-off on these sampling dates amounted to about 22—25 $dm^3\ sec^{-1}\ km^{-2}$ in the upper and lower reaches of the Nida and constituted 22—25% of the total run-off, calculated for all the sampling dates over the two year study period.

The trophic level and the degree of biodegradation was determined on the basis of the quantity of oxygen consumed by the bacteria in cultures kept in the water of particular stations, enriched with asparagine. Asparagine was used since aminoacids occur commonly in aquatic environments (Hobbie et al. 1968, Hall et al. 1970, Litchfield, Prescott 1970) and the requirement for asparagine and its uptake by bacteria in the processes of growth and development are large (Fonden 1969, Halvorson 1972, Godlewska-Lipowa 1974, Donderski 1983).

The results obtained confirm the increase in eutrophication of the water in the lower part of the catchment area on the Nida, where the production of biomass in cultures with asparagine was about 60—70% higher than in the upper part. These results are in agreement with those from Godlewska-Lipowa (1974, 1979), who found in the Masurian lakes an increase in the numbers and biomass of bacteria and in the coefficient of heterotrophic activity, in relation to asparagine and some other substrates with an increase in the trophic level of the water.

During the study period, distinct fluctuations in the production of bacterial biomass were found in the cultures with asparagine and its

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marked increase, mainly in summer but also in autumn. On the one hand, this confirms the relationship indicating an increase in the intensity of enzymatic processes with the rise in water temperature in summer (Christian, Wiebe 1974, Innis, Young 1977, Williams, Crawford 1983, Styevnyeva, Sudakova 1984); on the other hand, it indicates that the degree to which bacteria utilize an added substrate varies, depending on the time of year, which is in turn connected with the presence of specific groups with varying trophic requirements (Donderski 1983).

The greater amounts of energy used for bioaccumulation, and also for biodegradation in the water of the lower part of the drainage basin of the Nida, unequivocally confirm that the trophic level of the water increases with the course of the river. This is also indicated by the value of Schrödinger's coefficient (Odum 1982), which describes the "energetic maintenance costs" of the associations studied and indicates that the amount of energy per unit of bacterial biomass is much smaller in the lower part of the catchment area then in the Czarna and Biała Nida. Such an inference is supported by the coefficient of heterotrophic activity, A/K, which was found to assume fairly uniform and lower values in the water of upper part of the catchment area (Stations 1 and 2) and higher ones for the Nida at Station 4. In the case of the eutrophicated waters of the Nida at Station 3, the fact that the value of A/K remains at the same level as in Czarna and Biała Nida may indicate roughly equal trophic levels of the water. However, one should bear in mind that it was at this station that bioaccumulation and especially the biodegradation of organic matter in the water was the greatest, and clearly suggesting the highest quantities of nutrients in the water, as compared with the remaining stations. Besides, the value of the A/K coefficient of heterotrophic activity lower than at Station 4 may reflect the reduced assimilation of asparagine, resulting from the inflow of other substrates together with the sewage from Pińczów, of which the former may be more readily utilized by the bacteria. A similar decline in bacterial activity with respect to several substances of differing chemical structure, including asparagine, was found by Godlewska-Lipowa (1974) in Lake Nidzkie (Mazurian Lake District), polluted with wastes from a cellulose factory.

The results obtained fully document the increase in the trophic level of the water with the course of the River Nida, and allow division of the stations studied into two groups. The first consists of Stations 1 and 2, situated on the Czarna Nida and the Biała Nida, along sections where a low nutrient supply and high permeability characterize the soils near the banks, and the second comprises Stations 3 and 4, localized on the Nida and surrounded by soils of low permeability and abounding in nutrients.

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It appears that the varying character of the soils in the lower and upper parts of the drainage basin might contribute to smaller or larger fluctuations in the trophic level of the water, which were evident in periods of increased surface run-off. It is common knowledge that the use of land for agricultural purposes (fertilization, vegetation cover) and the properties of the soils themselves, connected with the abundance of nutrients, soil capacity, and permeability, are factors which affect the quantity and quality of the surface run-off, thus affecting the supply of nutrients in the water of rivers or water bodies (Margowski 1976).

It should be stressed, however, that the increase in nutrients in the lower part of the drainage basin of the Nida was affected not only by surface run-off but also by many other factors connected with the differences in the natural environment of particular sections of the river — such as the geological structure of the substratum, hydrological conditions, and pollution — and the cumulative effect of the material carried with the current from its upper reaches.

6. Polish summary

Aktywność flory bakteryjnej w rzece Nidzie na odcinkach różniących się podłożem i rodzajem przybrzeżnych gleb (Polska Południowa)

W latach 1982—1984 przeprowadzono badania nad aktywnością bakterii wód Czarnej Nidy (st. 1), Białej Nidy (st. 2) i Nidy (st. 3, 4) (ryc. 1).

Celem badań było określenie aktywności bakterii pod wpływem substancji spływających z otaczającego terenu, na odcinkach rzek różniących się typem podłoża geologicznego i przybrzeżnymi glebami (tabela I).

Aktywność mikroflory bakteryjnej określono przez porównanie ubytków tlenu w hodowlach prowadzonych na wodzie naturalnej (nie wzbogaconej) i hodowlach z dodatkiem asparaginy (100 μM dm^{-3} wody). Z ilości tlenu pobranego przez bakterie wyliczono produkcję ich biomasy w C mg dm^{-3} 24 h^{-1}. Zawartość biomasy podano w mg s.m. dm^{-3} wody. Obliczono współczynnik Schrödingera (R/B), wyrażający stosunek respiracji do biomasy i określający wydatki energetyczne na jednostkę biomasy oraz współczynnik heterotroficznej aktywności (A/K), będący ilorazem produkcji biomasy w hodowlach z asparaginą i w hodowlach kontrolnych, prowadzonych na wodzie naturalnej.

Wyrównaną produkcję biomasy bakterii w wodzie naturalnej stwierdzono na st. 1, 2, 4 oraz o 50% wyższą na st. 3 (ryc. 2A). W wodzie wzbogaconej asparaginą wykazano znacznie większą średnią produkcję biomasy (ryc. 2B).

W poszczególnych sezonach badawczych stwierdzono wahania produkcji biomasy w wodzie naturalnej i z dodatkiem asparaginy, co mogło być związane ze wzmożonym powierzchniowym spływem na przełomie lat 1982—1983 bądź z wyższą temperaturą wody w okresie letnim, która w istotny sposób determinuje intensywność procesów enzymatycznych (tabela II).

Wzrost produkcji biomasy w hodowlach z asparaginą o około 60—70% na st. 3, 4 (ryc. 2B) oraz stwierdzone znacznie większe ilości energii zakumulowanej w biomasie bakterii przemawiają za wyższym poziomem troficznym wód w obrębie tych stanowisk

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(ryc. 3). Wskazują na to także wartości współczynników R/B i A/K, mówiące o spadku „energetycznych kosztów utrzymania" i podwyższeniu heterotroficznej aktywności bakterii wraz ze wzrostem trofii środowiska w dolnym odcinku Nidy (ryc. 3).

Na podstawie uzyskanych danych badane stanowiska można podzielić na dwie grupy. Pierwsza ze stanowiskami na Czarnej (st. 1) i Białej Nidzie (st. 2), z przybrzeżnymi glebami o małej zasobności pokarmowej i dużej przepuszczalności oraz drugą ze stanowiskami na Nidzie (st. 3, 4), gdzie przybrzeżne gleby odznaczają się dużą zasobnością pokarmową i małą przepuszczalnością. Jednak należy podkreślić, że wzrost poziomu troficznego wody w dolnej części zlewni Nidy kształtowały nie tylko spływy powierzchniowe, ale także wiele innych czynników związanych z różnicami w środowisku przyrodniczym poszczególnych odcinków rzek oraz sumującym się działaniem wpływu materiału niesionego z nurtem z górnych partii rzeki.

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