

Auto- and heterotrophic picoplankton in a lowland river (Warta River, Poland)

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Abstract

The structure of plankton in a lowland river was studied with a particular focus on autotrophic as well as heterotrophic picoplankton cells. Water samples were collected in the middle, meridional section of the Warta River in 2005. Heterotrophic picoplankton (HPP) were represented by bacteria varying in size and form, but mainly by cocci and rods. HPP abundance ranged from 3.5×10^6 to 9.5×10^6 cells ml⁻¹. Autotrophic picoplankton (APP) densities were considerably lower and ranged from 1.2×10^3 to 92×10^3 cells ml⁻¹. They were represented by prokaryotes (picocyanobacteria) and eukaryotes (small chlorophytes). Both components occurred together but eukaryotic APP usually prevailed in terms of biomass. In terms of biomass, APP seemed to play a minor role in this ecosystem in comparison with heterotrophic picoplankton.

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INTRODUCTION

The riverine plankton varies in taxonomic composition and size structure, but for many years research focused only on larger organisms, i.e., nanoplankton and microplankton. Mainly due to methodological difficulties, small-sized organisms were formerly ignored or regarded as detritus. Only after the popularization of epifluorescence microscopy in the 1980s were many reports on picoplankton published. These small cells dominate numerically over larger organisms and can be important contributors to both photosynthetic and heterotrophic biomass and production. Many studies have focused on the importance of picoplankton cells, mainly in lakes (Stockner and Antia 1986, Sondergaard 1991, Weisse 1993, Stockner et al. 2000), coastal areas (Gomes et al. 1998, Carvalho et al. 2004), and estuarine systems (Moreira-Turcq et al. 2001), but only a few studies have been conducted in rivers (Marshall and Affronti 1992, Szelał-Wasielewska 2004, Gołdyn and Szelał-Wasielewska 2005).

The goal of the present study was to analyze the abundance and biomass of bacterioplankton and autotrophic picoplankton communities in a lowland river focusing on picoplankton cells within the bacterioplankton and their morphotypes. The chosen ecosystem was the Warta, a eutrophic river in mid western Poland.

MATERIALS AND METHODS

Water samples were collected from the current 0.2 m below the water surface 16 times between July and December 2005 (every 1–2 weeks) at one sampling site in the middle section of the Warta River (Wielkopolska Lake District, Poznań Water-Gap). The samples were preserved with buffered glutaraldehyde to a final concentration of 1%, and stored in a refrigerator at 4°C. Microscopic analyses were performed in the laboratory after the water samples had been concentrated on black Nuclepore filters (pore size 0.2 µm) and stained with DAPI (4', 6'-diamidino-2-phenylindole). The filters were next examined with cell autofluorescence and secondary fluorescence (Porter and Feig 1980, MacIsaac and Stockner 1993) under an epifluorescence microscope (Olympus BX-60) at a magnification of ×1500 with green, blue, and ultraviolet light excitation. Four morphotypes of bacterioplankton (cocci, rods, curved, and filamentous cells) were identified. In addition, two size groups were distinguished: smaller than 2 µm and larger than 2 µm. Autotrophic picoplankton were divided into prokaryotic and eukaryotic cells based on autofluorescence color, cell shape, and size. Cell abundance was expressed per ml, and cell volume was estimated based on measurements of their size with an



Fig. 1. Location of the sampling station on the Warta River in Poznań.

2006) at a station located several km upstream from the currently investigated section of the river, the max. concentrations of total nitrogen, total phosphorus and total organic carbon were as high as $6.9 \text{ mg l}^{-1} \text{ N}$, $0.5 \text{ mg l}^{-1} \text{ P}$ and $13.1 \text{ mg l}^{-1} \text{ C}$, respectively. Moreover, concentrations of chlorophyll *a* were very high (up to $187 \text{ } \mu\text{g l}^{-1}$, mean $70.1 \text{ } \mu\text{g l}^{-1}$), indicating the eutrophic state of the river.

RESULTS

Both bacterioplankton abundance and biomass were largely dominated by cells smaller than $2 \text{ } \mu\text{m}$, i.e., heterotrophic picoplankton (HPP). Their mean contribution to total bacterioplankton abundance and biomass was an average of 98.8% and 87%, respectively. HPP abundance ranged between 3.54×10^6 and $9.52 \times 10^6 \text{ cells ml}^{-1}$ (mean $5.89 \times 10^6 \text{ cells ml}^{-1}$), and biomass between 0.11 and 0.37 mg l^{-1} (mean 0.22 mg l^{-1}) (Table 1). The patterns of changes in HPP abundance and biomass during the study period were similar. Sigmoid variations in HPP biomass were observed during this study, but the successive biomass peaks were lower and lower: 0.35 mg l^{-1} in early September, 0.23 mg l^{-1} in early October, and 0.17 mg l^{-1} in late November and early December (Fig. 2). The contribution of HPP to total picoplankton abundance ranged from

Olympus image analysis system. Their biomass was then expressed as wet weight. Statistical analyses were done with Statistica 5.5 software.

The Warta River, the major right-bank tributary of the Oder River, is the third longest river in Poland and its basin covers approximately one-sixth of the area of the country. It flows through the Wielkopolska Region (mid western Poland). Its length is 808 km but the present study was restricted to the Poznań section of the river (Fig. 1). The river waters within the city of Poznań are very fertile; in 2005 according to the Provincial Inspectorate of Environmental Protection (Tybiszewska et al.

Table 1

Abundance and biomass of autotrophic (APP) and heterotrophic (HPP) picoplankton, and contribution (%) of HPP to total picoplankton abundance and biomass

Date	Picoplankton abundance (cells ml ⁻¹)			Picoplankton biomass (mg l ⁻¹)		
	APP	HPP	% of HPP	APP	HPP	% of HPP
12.07	92 341	4 953 248	98.17	0.0548	0.1832	76.97
31.07	9 303	7 034 263	99.87	0.0091	0.2710	96.75
07.08	34 592	7 992 697	99.57	0.0932	0.3627	79.56
14.08	11 266	9 524 228	99.88	0.0261	0.3697	93.41
21.08	15 589	6 984 422	99.78	0.0357	0.2753	88.52
28.08	8 651	3 541 942	99.76	0.0280	0.1306	82.34
04.09	4 332	8 492 228	99.95	0.0127	0.3544	96.54
11.09	1 581	7 305 547	99.98	0.0017	0.2987	99.43
18.09	4 443	5 624 931	99.92	0.0093	0.1946	95.44
25.09	2 990	5 168 988	99.94	0.0052	0.1780	97.16
07.10	1 207	6 261 622	99.98	0.0025	0.2312	98.93
23.10	2 323	5 123 646	99.95	0.0038	0.1575	97.64
04.11	3 818	3 719 799	99.90	0.0056	0.1228	95.64
18.11	2 906	4 280 469	99.93	0.0025	0.1676	98.53
06.12	2 545	4 307 490	99.94	0.0027	0.1541	98.28
19.12	3 592	3 906 323	99.91	0.0040	0.1060	96.36
mean	12 592	5 888 865	99.78	0.0186	0.2223	93.22

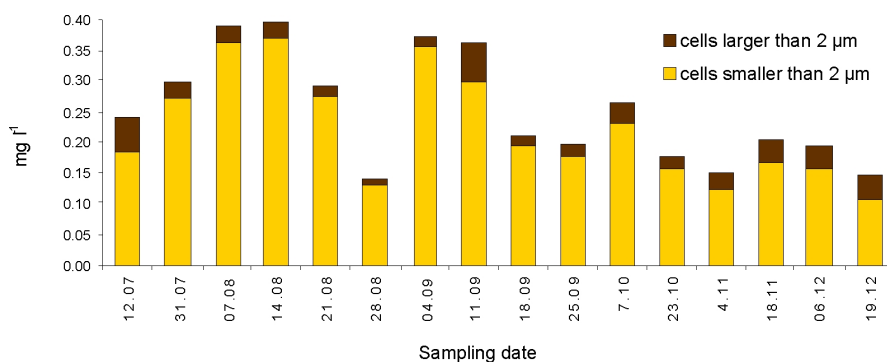


Fig. 2. Biomass proportions of heterotrophic cells smaller and larger than 2 µm.

98.2% to 99.9% (average 99.8%), and biomass reached an average of 93% and never fell below 75% (Table 1). Among the HPP morphotypes observed, cocci dominated in all water samples. Their mean contribution to total HPP abundance was 57.7% (45.1–82%). The mean contributions of rods and curved cells were 32.5% (11.5–42.8%) and 9.8% (4.6–21.1%), respectively. The mean cell volume of HPP cells was $0.037 \mu\text{m}^3$ and ranged between 0.027 and $0.045 \mu\text{m}^3$. In HPP biomass, cocci again were the most important morphotype, as their mean contribution to total picobacterioplankton biomass was 51.2% (32.4–80%). The mean contributions of rods and curved cells were 38.4% (10.8–49.3%) and 10.4% (1.8–21.3%), respectively (Fig. 3).

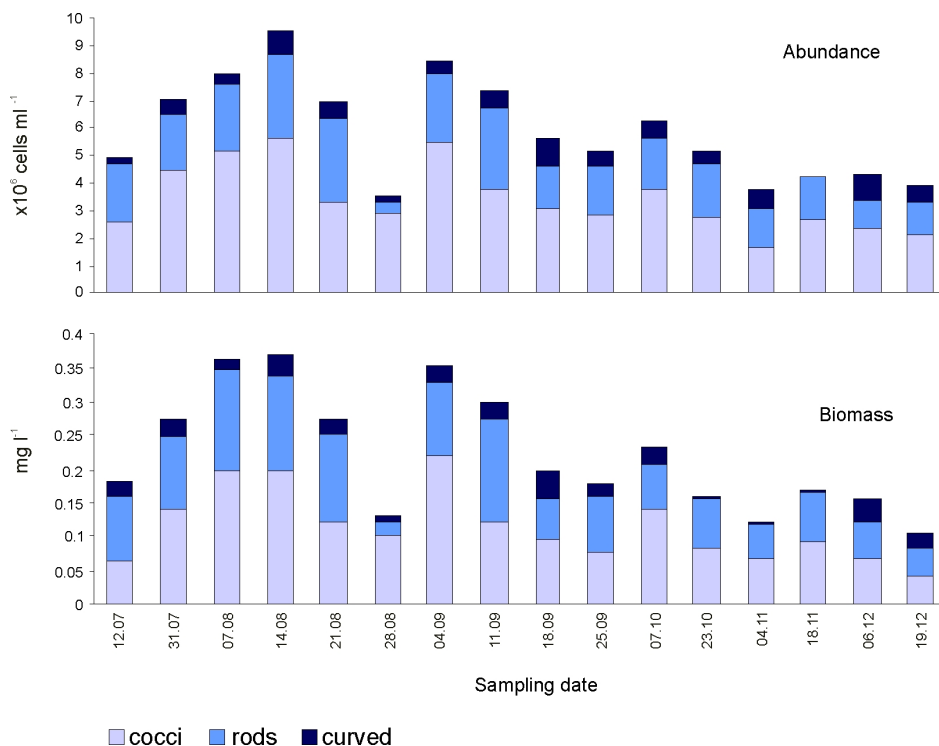


Fig. 3. Abundance and biomass of morphotypes of heterotrophic picoplankton (HPP).

Bacteria cells larger than $2 \mu\text{m}$ were less abundant than HPP, and filamentous bacteria occurred within this size fraction. This morphological form had a considerable effect on total bacterioplankton biomass on two sampling dates, i.e., in mid July (14.1%) and mid September (12.0%), but on average its

contribution reached only 3.3%. Among filamentous forms, the most abundant were the cells that did not exceed 25 μm in length and 0.3 μm in width. Only in mid September did some longer filamentous cells appear which reached up to 450 μm in length.

The total abundance of heterotrophic bacteria ranged from 3.56×10^6 to 9.60×10^6 cells ml^{-1} (mean 5.56×10^6 cells ml^{-1}) at a biomass of between 0.14 and 0.4 mg l^{-1} . The abundance and biomass of bacterioplankton were significantly and positively correlated with water temperature (for both $r=0.56$ and $p=0.023$). From mid July until mid September, when mean water temperature exceeded 18°C, the mean bacterioplankton biomass (0.3 mg l^{-1}) was about one third higher than in the period from mid September to mid December, when water temperature fell below 18°C (0.19 mg l^{-1}).

Throughout the study period autotrophic picoplankton (APP) abundance fluctuated from 1.2×10^3 to 92.3×10^3 cells ml^{-1} and APP biomass from 0.002 mg l^{-1} to 0.093 mg l^{-1} . The highest values of APP abundance and biomass were noted in mid July and early August. Changes in abundance were clearly visible from July to September, but only low values of up to about 5×10^3 cells ml^{-1} were observed until the end of 2005 (Table 1). Total APP abundance and biomass were not significantly correlated with water temperature ($r=0.40$, $p=0.11$, and $r=0.45$, $p=0.08$, respectively).

Within APP, both prokaryotic (cyanobacteria) and eukaryotic (chlorophyte) cells were noted. In terms of abundance, picocyanobacteria dominated more often, but in terms of biomass picoeukaryotes usually prevailed due to their larger cell size. The mean abundance of picocyanobacteria was 8.7×10^3 cells ml^{-1} and their contribution to total APP abundance ranged from 17 to 99% (mean 53%), while the contribution to total APP biomass was from 3 to 97% (mean 24%). The mean biomass of picocyanobacteria was low at 0.005 mg l^{-1} . Picocyanobacteria included single-celled and colony-forming species. The former picocyanobacteria often dominated during the study period both in terms of abundance and biomass (Fig. 4). Their contribution to the total abundance of picocyanobacteria ranged from 29 to 95% (mean 68%), while colony-forming picocyanobacteria cells comprised from 0 to 85% (mean 32%).

Eukaryotic APP were often more numerous than picocyanobacteria, particularly in August and September. Their abundance fluctuated from 0.2×10^3 to 23.9×10^3 cells ml^{-1} (mean 3.9×10^3 cells ml^{-1}). Picoeukaryotes usually accounted for a smaller proportion of total APP abundance (mean 47%, range 1–83%) than of total APP biomass (mean 76%, range 3–97%). The biomass of picoeukaryotes varied from 0.001 to 0.088 mg l^{-1} (mean 0.014 mg l^{-1}) and was higher than the biomass of picocyanobacteria in 15 out of 16 sampling sessions (Fig. 4).

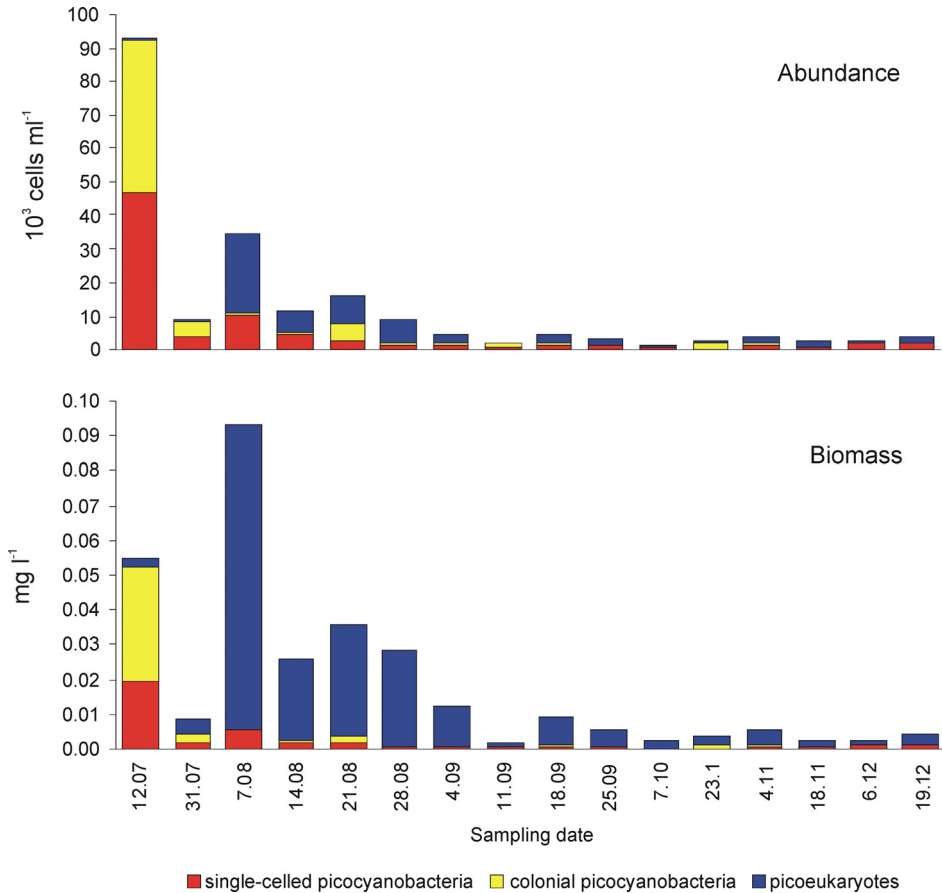


Fig. 4. Abundance and biomass of autotrophic picoplankton (APP) groups: single-celled picocyanobacteria, colonial picocyanobacteria and picoeukaryotes.

DISCUSSION

The density of heterotrophic plankton bacteria varied significantly depending on ecosystem type and trophic status. In general, total bacterioplankton abundance increases from the more oligotrophic open oceans toward coastal areas and estuaries. The mean bacterioplankton abundance in the Warta River of 6.0×10^6 cells ml⁻¹ fell within the range of values reported by Sorokin (1999) for eutrophic environments. Higher mean values of bacterioplankton abundance were recorded in the nearby Cybina River (a tributary of the Warta), at its inflow into the Antoninek (10.9×10^6 cells ml⁻¹)

and Maltański (8.3×10^6 cells ml⁻¹) reservoirs (Gołdyn and Szelaż-Wasielewska 2005). A much lower value (1.59×10^6 cells ml⁻¹) was reported by Zieliński and Leśniewska (2006) for the Narew River at its inflow into the Siemianówka Reservoir.

Maximum densities of bacteria are generally much higher than mean values for some research periods. In the Warta the highest value (9.6×10^6 cells ml⁻¹) was about three fold lower than in the Cybina at its inflow into the Antoninek Reservoir (Gołdyn, Szelaż-Wasielewska 2005) and about two fold lower than in the Warnow River (Freese et al. 2006). With such a high density, the bacterioplankton might serve as a single source of nutrition for most aquatic animals, including ciliates (Sorokin 1999).

One of the environmental factors that may have affected the bacterioplankton community in the Warta was water temperature. Some investigators (Freese et al. 2006, Zieliński and Leśniewska 2006) observed a positive relationship between water temperature and the abundance of bacteria. The present study of the Warta confirms that this correlation is significant, although only the second half of the year was it taken into account. The Warta River carries large amounts of suspended solids and chlorophyll *a*, partly originating from the fertile Jeziorsko Reservoir, which is located upstream, so bacterioplankton growth in the river is influenced by the large amounts of organic matter from that reservoir.

The APP abundance in the Warta during the sampling period in 2005 was generally low (mean 12.6×10^3 cells ml⁻¹) and never exceeded the value of 10^5 cells ml⁻¹, which was recorded in an earlier study of this river (Szelaż-Wasielewska 2004). That study was carried out in 2003 and mean APP abundance was almost three times as high then as in 2005. A similar mean value (13.6×10^3 cells ml⁻¹) was recorded in the waters of the Cybina River above a cascade of small reservoirs (Gołdyn and Szelaż-Wasielewska 2005). In contrast, Marshall and Affronti (1992) found a major growth of APP and mean densities of about 10^5 to 10^6 cells ml⁻¹ in three major tributaries of the Chesapeake Bay. The total APP density was also higher in the riverine zone of the Sardis Reservoir (southeastern U.S.) at a mean of 4.7×10^5 cells ml⁻¹ (Rhew and Ochs 2000). In 2005, APP density in the Warta River, as in 2003, declined along with seasonal drops in water temperature (Szelaż-Wasielewska 2004), but this relationship was not statistically significant. The biomass of APP was also low, which suggests that the smallest autotrophic cells do not find suitable conditions for development in polluted aquatic ecosystems, like the Warta. This may be associated with the sensitivity of APP to eutrophication and contamination, especially to heavy metals (Severn et al. 1989, Weisse 1991, Weisse and Mindl 2002).

Throughout the study period in the Warta River, HPP prevailed numerically over APP. Mean HPP abundance was about 470 times as high as that of APP. The maximum densities of the two groups were recorded in different periods; that of APP was noted at the beginning of the study period, while for HPP it was noted in the second month of the study in August. As reported by Stockner and Antia (1986), the abundance of picoplankton algae accounts for 5-20% of the total picoplankton abundance. In waters of the Warta, the difference in abundance between APP and HPP was even greater, as APP abundance was always lower than 2% of total picoplankton abundance. Similarly low values of abundance, i.e. APP below 1% of total picoplankton, were recorded in the Cybina at its inflow to the Antoninek and Maltański reservoirs (Gołdyn and Szeląg-Wasielewska 2005). In contrast, the contribution of APP in marine ecosystems was higher, as shown by Żmuda (2005), who found that autotrophic bacteria comprised on average 13.5% of total bacterioplankton biomass. The appearance of maximum abundance of APP and HPP in different periods may attest to differences in their environmental requirements, the ability to use food resources and/or sensitivity to the pressure of consumers.

CONCLUSIONS

The results of 16 sampling sessions in 2005 were used to analyze bacterioplankton and autotrophic picoplankton (APP) in the Warta, a eutrophic river in mid western Poland. The majority of picoplankton biomass was contributed by heterotrophic cells (HPP), which suggests that this component of the microbial loop is more important to the carbon and nutrient dynamics of lowland rivers than APP. HPP growth is facilitated by high concentrations of organic matter in water, while the low contribution of autotrophic cells can result from the turbulent flow of water and unfavorable light conditions for phototrophs.

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