Original Research

Succession of Periphyton Developing on Artificial Substrate Immersed in Polysaprobic Wastewater Reservoir

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Abstract

The succession of periphyton developing on artificial substrate immersed in a polysaprobic wastewater reservoir was studied during a two-year period. Three stages in the periphyton development: initial, intermediate and mature, were observed. The initial stage was characterized by domination of euglenophytes; *Carchesium polypinum* was dominant during the intermediate stage, while diatoms and chrysophyceans dominated the mature stage. However, the most characteristic component of periphyton consisted of detritus that contributed more than 50% (by volume). The periphyton development dynamics in the second year of the experiment, when that formation was already firmly established on the polyethylene sheets, was quantitatively similar to the process of periphyton development in the first year of substrate exposure.

Keywords: periphyton, ecological succession, artificial substrate, experiment

Introduction

There is still no commonly accepted definition of periphyton. According to the definition applied most often, the *sensu stricto* periphyton is an assemblage of plant and animal species that colonize various types of substrates in aquatic environments. Periphyton includes organisms that are attached directly to the substrate as well as those which move freely among them [1- 3]. However, in a broader sense, periphyton encompasses not only organisms, but also detritus (organic remains) and calcium carbonate. These are constant and often abundant periphyton components [3], for which reason both detritus and calcium carbonate were included in the present study.

Periphyton develops on both natural and artificial substrates. Glass plates are a type of artificial substrate used very frequently for research purposes [4, 5]. How-

ever, large-scale experiments, particularly those designed as biomanipulative operations for studying effects on periphyton on water purification, using polyethylene sheets seem to be a more advantageous approach [6-8]. Polyethylene properties such as flexibility and low specific weight make it possible to use large-sized sheets.

A survey of the most recent literature dealing with periphyton shows a growing interest in peryphiton chemistry, viewed from the standpoint of water self-purification. Numerous papers have addressed periphyton retention of nutrients [7, 9-12] and toxicants [13] as affected by factors such as light regime [12, 14], desiccation [12], and flow [15]. On the other hand, periphyton succession-related processes have been treated very selectively. Langis et al. [16] presented qualitative characteristics of a biofilm at an early stage of its development. Wołowski [17] studied taxonomic composition of algae occurring in an uncovered trickling filter of a sewage treatment plant. Lukin [18] identified algal groups dominating individual stages of periphyton community development. The pres-

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ent paper aimed to follow the periphyton succession on an artificial substrate deployed in a polysaprobic reservoir as a manipulation-based ecosystem-scale experiment [19]. Two aspects of the problem were analyzed: the establishment of periphyton in a reservoir devoid of it (year 1) and the further development of the periphytic community in the reservoir (year 2).

Knowledge of quantitative characteristics of periphyton is important because it is necessary for the assessment of actual effects of periphyton on self-purification processes in water bodies. Jöbgen et al. [7] studied phosphorus removal from eutrophic lakes using periphyton on submerged artificial substrata. The role of periphyton in self-purification was assessed from changes in chlorophyll as an estimate of algal biomass and total phosphorus concentrations. Sabater et al. [20] described the effects of biological factors on the efficiency of river biofilms in improving water quality. Among the biological factors they considered were community composition, biofilm structure, and the presence of grazers were found to be responsible for variations in the efficiency of selfdepuration. Szlauer and Szlauer-Łukaszewska [21] and Szlauer [22] studied the effect of periphyton on polluted waters.

Methods

Study Area

The experiment was carried out in a 65 m long, 20 m wide retention pond fed from the municipal and domestic sewage system. In terms of water quality, the reservoir was classified as polysaprobic (total suspended particulates: 67.8 mg/dm³; BOD₅: 27 mg/dm³; dissolved oxygen content: 0.4 mg/dm³; nitrite nitrogen content: 0.074 mg/dm³; total manganese content: 0.08 mg/dm³; total copper content: 0.1692 mg/dm³; lead content: 0.11 mg/dm³; volatile phenol content: 0.16 mg/dm³; chlorophyll *a* content: 536.5 μ g/dm³). It was characterized by variable environmental conditions, including wide fluctuations of water level (depth changing from 1.6 to 0.5 m) and flow rate. The pond was frequently affected by unknown substances causing a complete or almost complete disappearance of planktonic crustaceans and the presence of a thick sapropel layer on the bottom. Oxygen deficiency, particularly at the sediment-water interface, was frequent as well. Due to the absence of a suitable substrate (macrophytes), a natural periphyton formation was absent in the reservoir. The sapropelic bottom sediment ruled out the presence of benthic organisms [1].

The experiment was carried out over two years. It began by deploying a polyethylene sheet (1.03.98). A similar substrate was immersed in the reservoir at the beginning of year 2 (26.03.99). The sheets were placed vertically in the water column, using a system of floats and weights (Fig. 1).



Fig.1. Polyethylene sheets were immersed vertically in the water column with the aid of floats and sinkers.

Sampling Methods

Periphyton samples were collected from segments of the polyethylene sheets, retrieved from the reservoir. Two samples were taken during each sampling event, from different parts of sheets. Samples were taken 1 week after deployment and then at 2–3 week intervals through 4/28/1998 and then sampled 12 additional times at approximately monthly intervals through 9/1/1999.

Fresh periphyton samples were delivered to the laboratory. Once there, the periphyton was scraped from the polyethylene sheets. The Protozoa and Euglenophyceae were examined live and identified. Protozoan nuclei were stained with methylene green before identification. The samples were then preserved with 4% formaldehyde. Next, the area of a polyethylene segment from which the sample originated was calculated, while the periphyton sample was left in a measuring cylinder to sediment for 48 h, whereupon the sample volume was measured. The remaining taxa of algae and invertebrates were identfied from the formalin-preserved samples. Permanent slides for diatom identification were prepared in a special way: first, the sample was treated with 10% hydrochloric acid to remove carbonates; then, distilled water was added and the sample was left for 12 hours, whereupon it was decanted. The procedure was repeated several times. Finally, the sample was boiled in 37% hydrogen peroxide to remove organic matter. Diatom slides were mounted in Naphrax.

The next step involved volume determination of periphyton microcomponents, following the method described by Szlauer [23]; the volume was expressed in cm³ per m² polyethylene sheet. Most organisms were identified to species, although some could be identified to a higher taxon only. Microcomponent volume calculations also included detritus and calcium carbonate. Once the volume determinations were over, the sample was sieved on a 0.5 mm mesh sieve to separate the macroscopic fraction. Usually, the macroscopic fraction is the first to be separated from the rest of the sample. In this study, a reversed order was applied to prevent some of the filamentous colonies of *Carchesium polypinum* (Protozoa) to pass into the macroscopic fraction. That fraction was rinsed and organisms were picked out from the sample residue under a stereo microscope. All the invertebrates were measured to calculate their volume, using appropriate conversion factors. As already indicated, volume rather than biomass was measured to ensure comparability between the abundance measures applied to macro- and microcomponents. The absolute volume of each component was scaled to a sheet surface area unit (m²).

Multivariate Analyses

The quantitative data obtained were processed using a set of multivariate techniques supplied by the PRIMER software (Plymouth Routines in Multivariate Ecological Research; [24]).

Similarity in periphyton structure between sampling periods and periphyton development stages was studied by means of the PRIMER's CLUSTER module, involving the Bray-Curtis similarity coefficient and double square root transformation of data, the latter applied to alleviate the effect of the wide variability in the original values. The similarity matrix was sorted with the group average strategy to produce a similarity dendrogram and to identify homogenous groups in it.

Multi-dimensional scaling (MDS) was applied to visualise the degree of dissimilarity, in a 2-dimensional plot, of sampling occasions within a multi-dimensional space; distances between individual sampling occasions in the plot and their arrangement reflect similarity or dissimilarity of corresponding periphyton assemblages. The MDS stress value indicates the degree with which a 2-dimensional diagram reflects the true relationships between the sampling dates.

PRIMER'S SIMPER procedure was applied to the data set to identify the taxa that contributed most to dissimilarities between periphyton assemblages on various sampling occasions compared.

Results

MDS plot (Fig. 2) grouped the sampling events into three areas. Two events (98/03/04 and 98/03/17) are situated away from the remaining ones and cover the period of the initial 17 days of the periphyton formation. The second area includes samples collected on 98/04/07 and 98/04/28 and covers the period from week 3 to week 11 of the periphyton development. The third area, situated close to the previous one, encompasses the remaining sampling occasions, i.e., the period from week 12 until the end of the experiment.

The similiarity dendrogram groups the sampling event in a way similar to that produced by the MDS plot: there are three clusters identified at the similarity level of about 50%. The first cluster covers the initial 17 days of the periphyton formation; the second cluster spans weeks 3 to 11, while the third cluster encompasses the period from week 12 until the termination of the experiment.

Both the MDS plot and the similarity dendrogram allowed the inference that the periphyton was being formed during three distinct stages:

- Stage 1: initial; the first 2 weeks of periphyton development on a newly immersed sheet (samples 4.03.98, 17.03.98);.
- Stage 2: intermediate; from week 3 to week 11 (samples 7.04.98., 28.04.98);
- Stage 3: mature; from week 12 until the end of experiment, assumed to be the mature, climax stage of the periphyton succession (samples 20.05.98-1.09.99).



Fig. 2. MDS plot (stress = 0.07) showing sampling occasions. Arrows shows time sequence expressing the succession.

Stages	Average dissimilarity	Taxa most responsible for the extent of dissimilarity indicated and their contribution to percent dissimilarity						
initial and intermediate	67.23	Carchesium polipinum (13.2), Chlorophyta (9.7), Bacillariophyceae (9.8)						
initial and mature	87.19	Bacillariophyceae (19.73), Chlorophyta (16.32), Chrysophyceae (12.37)						
Intermediate and mature	52.26	Chrysophyceae (7.6), Carchesium polipinum (7.1), Bacillariophyceae (6.45)						

Table 1. Degree of dissimilarity between stages of periphyton development for individual taxa.

The data reported in Table 1 identify those periphyton components that contributed most to dissimilarity and similarity between periphyton assemblages at different succession stages. The initial vs. intermediate stage comparison shows the protozoans, represented by *Carchesium polypinum*, and algae (green algae and diatoms) to be primarily responsible for differences between the two stages. The differences between the initial and the mature stage were mainly due to chrysophyceans, *C. polypinum*, and diatoms.

At the initial stage, the newly developing translucent, light-green periphyton, 18.5cm³/m² in volume, was primarily composed of detritus (11 cm³/m²) and euglenophytes (7.5 cm³/m²; Table 2). The euglenophytes consisted mostly of Euglena viridis fo. viridis, Euglena sp., and Lepocinclis aff. elongata. The intermediate stage was characterized by a change of colour to bottle green; the periphyton layer was thin and somewhat lumpy in texture. At this stage, the mean periphyton volume was 524 cm³/ m^2 , detritus being the major component (315 cm³/m²). The most important biotic components included C. polypinum (89 cm³/m²), diatoms (45 cm³/m²), and green algae (35 cm³/m²). The diatoms were dominated by Nitzschia palea and Gomphonema parvulum, while the chlorophytes were represented mainly by Chlorella sp. and Trochiscia prescotti. The euglenophyte volume was 10 cm³/m² (Table 2). During the intermediate stage, after nine weeks of exposure (in late April), the periphyton volume reached its maximum in year 1 (921 cm³/m²; Table 3).

The climax stage in the periphyton succession was characterized by the very dark bottle-green coloration and dense, lumpy texture of the assemblage. The mean periphyton volume was about 584 cm³/m² (Table 2). The major contributors included detritus (slightly more than 50%), followed by diatoms (about 107 cm^3/m^2 , i.e., 18% of the total volume and 40% of the volume of biotic components), spherical unidentified chrysophyceans (mean volume 87 cm³/m², i.e., 11 and 33%, respectively), and green algae which supplied less than 54 cm³/m², i.e., 9 and 20%, respectively. The diatoms were dominated by Nitzschia palea and Navicula seminulum, while the green algae were represented mainly by Scenedesmus acutus and Monoraphidium arcuatum. Much less voluminous were ciliates (mainly peritrichs) (1.6 and 4%, respectively) and the Cyanoprocaryota (0.8 and 1.5%, respectively; Table 2). Typical of the climax stage was a high contribution of calcium carbonate (6.4%). After 16 months of exposure (in late May), the periphyton volume attained its overall maximum (1068 cm^3/m^2 ; Table 3).

The total periphyton volumes in years 1 and 2, after identical durations of exposure, are compared in Fig. 4. After two weeks of exposure, the volume of periphyton exceeded 65 cm³/m² both in years 1 and 2. After about eight weeks of exposure, the volumes amounted to 730 and 560 cm³/m² in years 1 and 2, respectively. The volumes attained after 5 months of exposure in years 1 and 2 were 441 and 342.5 cm³/m², respectively.

The taxonomic composition of the periphyton varied considerably during both years 1 and 2. In year 1, 2 weeks after polyethylene sheet deployment, the dominant component was detritus, biotic components being somewhat less abundant. C. polypinum, a typical periphytic organism, appeared as the dominant biotic compound, followed by euglenophytes and by much less abundant cyanoprocaryotes, filamentous green-algae, and rotifers. In contrast, the periphyton in year 2 was dominated by calcium carbonate and detritus, biotic components occurring in low volumes only. Noteworthy is the much higher species richness in year 2, compared to year 1. The periphyton biotic components were in year 2 represented by small spherical chrysophyceans, green algae, ciliates, euglenophytes, and diatoms (Fig. 4).

Table 2. Average volumes of periphyton components (cm^3/m^2) at individual periphyton succession stages on the polyethylene sheets.

	Component volume								
	initial	intermediate	mature						
Cyanobacteria	0.0	3.9±3	4.1±3.7						
Euglenophyta	7.5	10.3±1.8	1.3±1.6						
Chrysophyceae	0.0	0.0±0	87.5±85						
Bacillariophy- ceae	0.0	45.0±59.5	107.3±65.2						
Chlorophyta	0.0	35.2±27.2	54.2±41.4						
C. polypinum	0.1	88.7±91.1	7.7±14.4						
Other Ciliata	0.004	20±23.8	2.652±3.8						
Other taxa	0.000	5.834±4.4	1.969±4						
CaCO3	0.0	0.0±0	39.2±14.6						
Detritus	10.9	314.6±218.1	278.5±120.3						
total	18.5	523.6	584.3						

Table 3. Average volumes of periphyton components (cm^3/m^2) on the polyethylene sheets 1.03.98- the beginning of exposure. Dissolved oxygen content in surface and near-bottom water layers (mg/dm^3) .

18	months	01.09.99.	10.1	0	146.2	135.3	8.1	0	0.4	0	38.3	368.1	706.6		0	0
15.7	months	21.05.99.	4.6	4.2	244.4	227.4	54.1	0	0	1.4	33.8	498.6	1069		3.2	1.6
13.7	months	19.04.99.	2	0	0	51.4	166.9	1.6	4.9	3.3	44.1	405.3	679.4		10.4	9.8
13.5	months	12.04.99.	8.4	0.9	167.8	37.5	27	7.7	10.6	0.3	24.2	332.8	617.2		14.2	6.8
12	months	8.03.99.	8.5	2.7	74.3	35.7	41.9	34.6	0.4	0.6	29.6	323.8	552.2		5.3	0
10	months	20.01.99.	6.3	2.4	139.9	46.4	72	38.2	1	0.6	26.9	278.1	611.8		2.1	0.4
8 months		6.11.98.	2.3	0.3	140.1	63.2	58.5	0	8.7	1.1	48.1	232	554.3		0.3	0.3
6.8	months	17.09.98.	0.4	0	49.5	160.9	37.3	0	0.7	7.6	75.4	185.2	517	u	0.9	0.7
5 months		4.08.98.	0	0	0	175.2	59.7	2.6	0	2.7	33.4	167.2	440.9	lved oxyge	2.1	3
4.7	months	22.07.98.	0	0	0	107.3	31.8	0	0.3	1.9	28.6	95	264.8	Disso	3.2	1.3
3.7	months	23.06.98.	2.8	3.4	0	139.8	38.8	0	2.1	2.1	48.7	177.2	415		3.3	2.7
2.5	months	20.05.98.	5.2	8	0	127.6	63	9.8	11.1	8	0	333.7	566.5		0	0
6.	weeks	28.04.98.	2.8	9.6	0	49.1	48.5	171.7	52.4	9.7	0	577.7	921.3		15.4	0
.9	weeks	7.04.98.	7.3	11.6	0	3.2	29.5	163.6	16.4	5.4	0	302.5	539.6		15.8	3.4
. 2	weeks	17.03.98.	0.5	11.8	0	0	0	9.9	0	0.3	0	44.4	6.99		14.1	2.1
4,	days	4.03.98.	0	7.5	0	0	0	0.1	0	0	0	10.9	18.5		13.5	1.4
Even courses	time		Cyanobac- teria	Euglenophyta	Chrysophy- ceae	Bacillari- ophyceae	Chlorophyta	C. polypinum	Other Ciliata	Other taxa	CaCO3	Detritus	Total		surface	bottom

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Fig. 3. A similarity dendrogram of periphyton assemblages on different sampling occasions.



Fig. 4. Volume of periphyton components in year 1 (I) and year 2 (II), sampled after identical periods of polyethylene sheet exposure.

After eight weeks of exposure in year 1, the periphyton was dominated by detritus and *C. polypinum*. The other periphytic components were considerably less abundant, but their species richness increased. Samples collected in year 2 showed domination by diatoms, followed by detritus, green algae, calcium carbonate, and small, spherical chrysophyceans. For comparison, after 8 weeks of exposure in year 2, the periphyton was dominated by diatoms and detritus (Fig. 4).

After 5 months of exposure both in year 1 and year 2, the volumes of detritus and calcium carbonate reached similar levels. However, the volume of biotic components and their taxonomic composition varied greatly. In year 1, dominant were diatoms and green algae, while cyanoprocaryotes, chrysophyceans, and diatoms dominated in year 2 (Fig. 4).

Fig. 5 shows periphyton components developing on the polyethylene sheets after various periods of exposure, that is it compares the newly forming periphyton with that present on the sheet exposed for more than a year. The compared samples (old and new periphyton) were always collected exactly at the same time. The major difference between the new and old periphyton was their respective volumes. The year 1 periphyton was always less voluminous, compared to the volume accumulated in year 2. For the samples collected in April, May, and September, the differences amounted to about 700, 800, and 400 cm^3/m^2 , respectively. The major factor accounting for the difference was the volume of detritus, which was always lower on freshly immersed polyethylene sheets, compared to that exposed for over one year. Volume of the remaining components was on the similar level at both substrates,

regardless of duration of exposure. Also regardless of duration of exposure, the most important groups among the biotic components were diatoms, chrysophyceans, and green algae (Fig. 5).

Discussion

The experiment demonstrated that deployment of a polyethylene sheet was a useful tool in creating the habitat for the formerly non-existent periphyton community, in the water reservoir. The initial stage of periphyton formation on the sheet was shown to involve a gradual increase in periphyton volume. While absent from the water body in year 1, the periphyton formed in that year continued to grow in year 2 on the sheets immersed the previous year. The biovolumes of the periphyton were similar in both years. However, there were distinct differences in the composition of the periphyton settling on the polyethylene sheets in the two years.

The absence of periphyton when the artificial substrate was immersed in the reservoir might be the major reason explaining the differences in sheet colonization patterns between this study and those reported in literature. In this work, the major biotic components were euglenophytes and *C. polypinum*. Euglenophytes, planktonic organisms, were reported from periphyton only sporadically. Wołowski [17] showed euglenophytes to be rare in periphyton in a sprinkled bed waste water treatment plant with sprinkling bed and to be more common in the rinsed sediment. Having studied the same type of sewage treatment plant, Wołowski [25] found euglenophytes to occur mainly in the foam gathering on the surface of the waste water. Euglenophytes are known to occur in periphyton



Fig. 5. A comparison of periphyton components developing on polyethylene sheet after various periods of exposure.

in highly polluted waters only [17, 26, 27]. The domination of euglenophytes revealed in the present study in the initial stage of periphyton formation was most probably caused by their settling out from the plankton [18]. The high volume of *C. polypinum* could have resulted from the ciliate's migration from decomposing tree leaves, deposited on the bottom of the reservoir studied.

The biofilm formation in sewage water was initiated very early, on day 1 to 4 [16, 17]. At that time, the periphyton consisted of bacteria and mucus. Bacteria, which can attach themselves to the substrate, form a biofilm which adsorbs or traps the suspended matter and algae from the water [28]. In the study described by Wołowski [17], the order of substrate colonisation was different from that observed in the present experiment. After eight days of exposure, only solitary cells of diatoms, cyanobacteria, and green algae have been identified [17]. Algal cell densities increased exponentially after 8 days of adding the algal suspension [28]. The species richness of periphyton assemblage studied by that author was increasing with time of exposure. The taxa identified by other authors [16, 17, 29], such as green algae, diatoms and cyanoprocaryotes appeared in substantial amounts in the periphyton studied as late as in week 8 of exposure.

In year 2, the situation in the reservoir changed considerably with respect to periphyton. The polyethylene sheets immersed a year earlier was supporting a fully developed periphyton community. The results obtained indicate that the substrate immersed in year 2 was colonized by periphytic organisms originating from the older sheet rather than from the water and decomposing tree leaves, as in the previous year. The periphyton assemblages present on the substrate immersed in year 2 and on that installed a year earlier were very similar in their taxonomic composition and proportions between biotic components. An identical finding was reported by Bohr [29], who found that the periphyton developing on glass slides exposed for 4 days was dominated by the species dominant on natural substrates; later on, the first colonisers grew in abundance and were supplemented by epiphytic algae, mainly diatoms, that were increasing in abundance. Wołowski [17] found peryphytic organisms, although less abundant on a new substrate, to be vital and including the taxa also present on older sprinkled beds. According to Pieczyńska [30], the major way the periphytic communities increase in abundance on a new substrate is via immigration of individuals, rather than through reproduction of the already existing ones. This, however, was the case of long-lived taxa. The short-lived and rapidly reproducing ones might have increased their abundance by intensive reproduction of the primarily settled individuals. Lukin [18], too, reported green algae to sediment from plankton to the periphiton.

In this study, the amount of detritus was substantially higher on the sheets exposed for more than a year, compared to that immersed in year 2, the difference persisting until the end of the experiment. A certain quantity of detritus is bound by and stored in periphyton due to the excreted mucus (EPS) [20, 31, 32]. Once bound, the detritus could be most probably kept in periphyton for at least one year, which can explain its higher amount in the substrate exposed for a longer time in the reservoir studied.

The periphyton volume reached its maximum in a newly immersed polyethylene sheet in year 2 in late April. Similarly, the highest volume on the substrate immersed the previous year was recorded in May. Some authors [32-35,] observed the periphyton to grow at the highest rate in spring. However, in numerous other studies [28, 36, 37], the highest periphyton biomass was recorded within July-September. The maximum periphyton volume observed in this study in April and May can be explained by favourable oxygen conditions at the time, no oxygen deficiency being recorded in the reservoir then.

As shown by this experiment, the periphyton needed about 12 weeks to reach its maximum volume on the newly colonized polyethylene sheets. This was also observed by Szlauer & Szlauer [6, 35]. The maximum volumes of periphyton microcomponents were very similar regardless of habitat type (e.g. a mesotrophic lake, a hypertrophic reservoir). The volume of about 1070 cm³/m² is probably the upper limit for a periphytic community developing on polyethylene sheets and consisting mostly of unicellular algae, protozoans, detritus, and calcium carbonate. In contrast, a periphytic community growing on a polyethylene sheet and dominated by colonies of bryozoans and *Cordylophora caspia* may attain as much as 4000 cm³/m² [38].

Having peaked in year 1, the periphyton volume decreased and remained at a stable level until the end of the experiment, the average volume amounting to about 550 cm³/m². This result is similar to data reported by Szlauer & Szlauer [6] from a mesotrophic lake. However, other authors [35, 39] recorded considerably lower volumes of periphyton microcomponents, which might have resulted from gastropod grazing. [7, 8, 18, 34, 40] associated the summer periphyton minimum with grazing by gastropods and chironomid larvae. It may thus be assumed that the microcomponent volume of 539cm3/m2, reported both in this present study and by Szlauer & Szlauer [6], is an average possible to be obtained on polyethylene sheets immersed in water bodies ranging from hypertrophic to mesotrophic, in the absence of strong gastropods or chironomid grazing. Periphyton of such volume is able to persist on the sheet surface due to specific features of the periphyton itself. The organisms inhabiting the biofilm are bound together by extracellular polysaccharides (EPS) [31], which may account for up to 70-95% of the periphyton dry weight [41].

Identification of constant periphyton taxa is crucial, as the components most important in terms of abundance and constancy occur throughout the year; their abundance only may vary [29], or they increase in abundance from spring until autumn [34]. That group included the components, both biotic diatoms, green algae) and abiotic (detritus and calcium carbonate) which persisted in the periphyton from the moment they first appeared until the end of the experiment. In year 1, the contribution of the constant components became stabilized following the peak periphyton volume (10 weeks of exposure). The array of constant periphyton microcomponents, be it dominants or subdominants, i.e. detritus, diatoms and green algae, was – in the reservoir studied – similar to that reported from lacustrine and riverine waters [6, 32, 35-37, 42]. On the other hand, proportions between the periphytic Cyanoprocaryota, chrysophyceans, and protozoans differed between water bodies studied. Chrysophyceans are rare in periphyton; their presence in periphytic communities was reported by Pizarro et al. [4] and Sekar et al. [32].

To determine the time necessary for the climax stage (mature stage) to be reached by periphyton, both quantitative (the periphyton volume) and qualitative (proportions between the permanent periphyton components) factors were taken into account. This is in agreement with observations of Roos [34] who found the periphytic climax to be characterized by the presence of a certain number of constant taxa, but not to be coincident with the maximum algal biomass.

In this study, the quantitative parameters of the climax stage were as follows:

- periphyton volume close to 550cm³/m² (this limit was determined for reasons described in this chapter); such volume was attained after 6 weeks;
- the following proportions of the constant components: about 50% supplied by detritus, about 18% contributed by diatoms, and about 9% supplied by green algae; those proportions were reached between weeks 10 and 14;

This is in agreement with results of the multivariate analyses which allowed suggesting the beginning of the mature stage at week 14. Data published by various authors show differences in this respect. According to [30], the time necessary for the climax to be reached is 16 to 20 weeks. Pieczyńska and Banaś [43] reported 8 (seldom 4) weeks, whereas the estimate of Szlauer & Szlauer [42] is 24 weeks. These differences, however, are not very substantial and, when averaged, produce a result similar to that of this study. The authors referred to might have used a much simpler method to determine the time the periphyton needed to become a stable community. It would be sufficient to compare the periphyton of the newly colonized substrates with that already existing. In this study, the periphyton had to be formed at first, so it was difficult to find the criteria with which to estimate the time necessary for the periphyton to become a stable community, particularly that the climax stage may be subjected to changes induced by succession or pollution [34].

In this study, polyethylene sheet modules were designed and their functioning tested. During the two years of exposure, the modules were neither destroyed nor displaced. Thus, modules can be successfully used to enhance self-purification processes in natural, polluted water bodies or in wastewater treatment reservoirs. Applicability of periphyton developing on a polyethylene sheet to water treatment was investigated and directly tested in a laboratory study by the present author [21] and by Szlauer [22]. Numerous other authors [6, 7, 35, 36, 38, 42] refer indirectly to the potential of the periphyton developing on plastic sheets to treat polluted waters.

Conclusions

- 1. The periphyton succession was proceeding in the following three stages:
 - initial stage: week 2 of periphyton development;
 - intermediate stage: weeks 3 to 11;
 - mature stage: week 12 until termination of the experiment.
- 2. The numerical values characterizing the climax stage, corresponding to the mature stage, were determined as follows:
 - periphyton volume close to 539cm³/m²;
 - proportions between constant components of 50% (detritus), about 18% (diatoms), and about 9% (green algae).
- When the periphyton is present in a water body, new substrates are colonized by periphytic organisms originating from that grouping and moving onto a new substrate.
- 4. When a reservoir lacks periphyton, it is formed initially by organisms originating from other ecological groupings, e.g. plankton. With time, typical periphytic forms become prevalent and dominant over those belonging to other ecological groupings. At the climax stage, the periphyton community shows a structure and composition typical of a water body under study.

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