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Research Article

**BLOOMING OF *APHANIZOMENON FLOS-AQUAE* IN  
THE URBAN POND**

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**Abstract**

Research on the mass occurrence of *Aphanizomenon flos-aquae* (L.) Ralfs ex Born. & Flah. was conducted in a shallow park pond in the city of Toruń from October 2003 until September 2004. A high ratio of  $N_{\text{tot}}:P_{\text{tot}}$  in the pond water did not limit the growth of the blue-green algae. The increased concentration of nitrates led to a reduction of the amount of heterocysts in the trichomes, whereas a deterioration of the habitat conditions and a high concentration of *A. flos-aquae* trichomes favoured the formation of akinetes.

## INTRODUCTION

*Aphanizomenon flos-aquae* (L.) Ralfs ex Born. & Flah. – one of the commonest species of this genus – has been the object of many taxonomic and ecological studies (Komárek & Kováčik 1989, Kohl *et al.* 1985, Kováčik & Holečková 1984). It blooms both in eutrophic and hypertrophic water bodies (Barbiero & Kann 1994, Pańczakowa & Szyszka 1986, Lin 1972, Pechar & Fott 1991, Jacobsen 1994), as well as in oligotrophic waters (Anagnostidis & Economou-Amilli 1988). Although *Aphanizomenon flos-aquae* is a characteristic species of summer lake phytoplankton, it sometimes blooms in winter, too (Jones 1979). According to a number of researchers (Burchardt 1998, Dokulil & Teubner 2000, Tsujimura *et al.* 2001, Takano & Hino 2000), a higher water temperature, the pH of 7.5, good light conditions and a low nitrogen-to-phosphorus ratio caused by an increased influx of phosphorus, all contribute to the domination of the blue-green algae. However, a single factor is hardly ever accountable for the massive emergence of *Cyanoprokaryota*. The long-term domination of the algae is a result of their joint influence.

There is little information on the occurrence of akinetes in natural basins or their initiative role in the growth of alga population. It has not been sufficiently substantiated if akinetes are a wintering phase in the development of algae, or if their springtime germination is to produce inoculum for subsequent growths (Jones 1979).

The aim of the study was to further understand the mechanisms behind the intense blooming of *Aphanizomenon flos-aquae* and the changing number of heterocysts and akinetes in the specific conditions of a small hypertrophic pond-like water body.

## MATERIALS AND METHODS

The physicochemical and phycological research was conducted in a city park pond (approx. 1.250 m<sup>2</sup>, 1.6 m deep) from October 2003 until September 2004. The water body is situated in a natural depression surrounded with trees. In summer, it is a dwelling place for birds. During the intensive blooming of blue-green alga, samples were taken every second day (from 22 October until 12 November), afterwards – twice a month. The input material was collected regularly between 10.00 and 11.00 o'clock in the morning. Samples for qualitative tests were collected with a phytoplankton net no. 25. The quantitative (that were not thickened) collected with a container directly from the bottom of the pond. The samples were fixed with 4% formalin or Lugol solution (IKI).

The biomass of the algae was determined based on the amount and volume of cells (volumetric method). The cells were counted in sedimentation chambers under an inverted microscope using 400x magnification. In order to calculate the volume of the trichome its average length of 100  $\mu\text{m}$  was assumed along with the average cell volume of 4  $\mu\text{m}$ . The number of heterocysts and akinetes in the trichomes was verified 30 times to capture its variability.

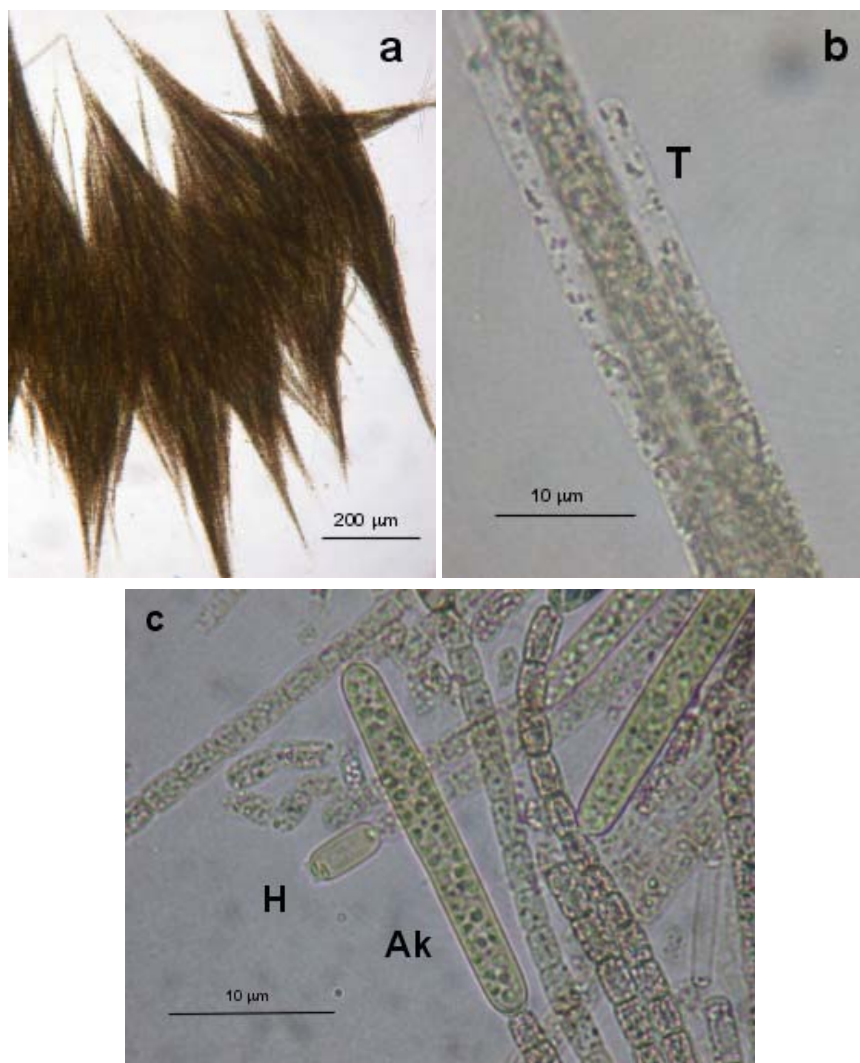
The concentration analyses of N-NO<sub>3</sub>, N-NO<sub>2</sub>, N-NH<sub>4</sub>, N<sub>k</sub>, N<sub>tot</sub>, P-PO<sub>4</sub>, and P<sub>tot</sub>, the content analysis of the dissolved oxygen and the concentration of chlorophyll were performed using standard hydrochemical methodology (Hermanowicz *et al.* 1976, Golachowska 1976, Nusch 1980). The measurements of temperature, pH indicator, and characteristic conductivity of the water were taken at each sampling.

## RESULTS

In the research period, the water temperature ranged from 2.7°C to 22.1°C. The water reaction was alkaline and the oxygen saturation exceeded 11 mg O<sub>2</sub> in 1 dm<sup>3</sup>. In December, the oxygen content in the water was observed to fall to 6.8 mg O<sub>2</sub> in 1 dm<sup>3</sup>. The high electrolytic conductivity of 416-1070  $\mu\text{S} \times \text{cm}^{-1}$  indicated a considerable presence of substances solved in the water.

During the research, a seasonal change was noticed in the diversity of composition of the pond plankton algae. Between 22 October and 3 November, the samples were dominated by *Aphanizomenon flos-aquae* which reached its maximum biomass in that time (113 mg in dm<sup>-3</sup>). From mid-November until the next July *Asterionella formosa* and *Scenedesmus sp. div.* co-dominated in the pond. In that period, *A. flos-aquae* disappeared from the plankton composition. In August and September 2004, the following blue-green algae co-dominated in the plankton: *Woronichinia compacta*, *Microcystis aeruginosa*, and *Planktothrix agardhii*, along with coccal green algae. At that point, trichomes of *Aphanizomenon* reappeared. The calculated amount of biomass of the algae in the water body under investigation was corresponding to the biomass of algae in fertile eutrophic lakes (Kentzer 2001). The concentration of chlorophyll *a* ranged from 2.2  $\mu\text{g}$  in dm<sup>-3</sup> (22 January) to 157  $\mu\text{g}$  in dm<sup>-3</sup> (22 October). The observed high concentration of chlorophyll *a* was positively correlated with the biomass of the algae. The pheophytin content at the sampling point reached approx. 70% of the total amount of active chlorophyll.

On the surface, the trichomes of *Aphanizomenon flos-aquae* formed characteristic sickle-shaped bundles (Fig. 1a). The vegetative cells in the



**Fig. 1.** *Aphanizomenon flos-aquae* from urban pond: **a** – Sickle-shaped aggregates of trichomes; **b** – Terminal cell (T); **c** – Heterocyst (H) and akinete (Ak).

trichomes are cylindrical or - occasionally - barrel-like (4.00-4.88 µm wide and 6.0-7.2 µm long) with gas vacuoles. The endings of the cell are elongated, narrow and highly vacuolized. In older trichomes, they become hialynized (Fig. 1b). The intercalary heterocysts, cylindrical in form (5.1-6.8 µm wide and 6.8-12.2 µm long), are usually single in the trichome (Fig. 1c). The amount of heterocysts in the trichome (Fig. 2) is variable. In autumn, when the

concentration of nitrates increased, a violent drop in the number of heterocysts was observed. In spring, when the nitrate content decreased and the concentration of  $N-NH_4$  was low ( $0.05 \text{ mg} \times \text{dm}^{-3}$ ), the occurrence of heterocysts in a trichome suddenly became more frequent. In the final stage of the research, some trichomes without heterocysts were observed. This process was accompanied by an increase of ammonia nitrogen concentration and a decrease of nitrate concentration.

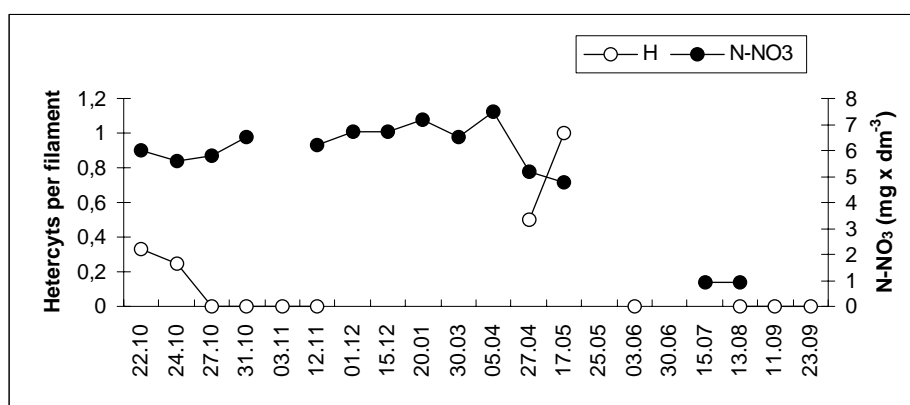


Fig. 2. Changes in the number of heterocysts in a trichome depending on the concentration of nitrates.

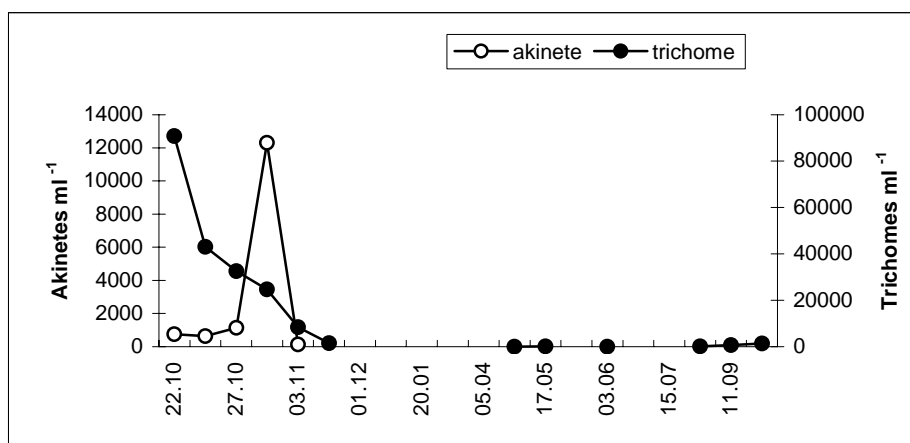
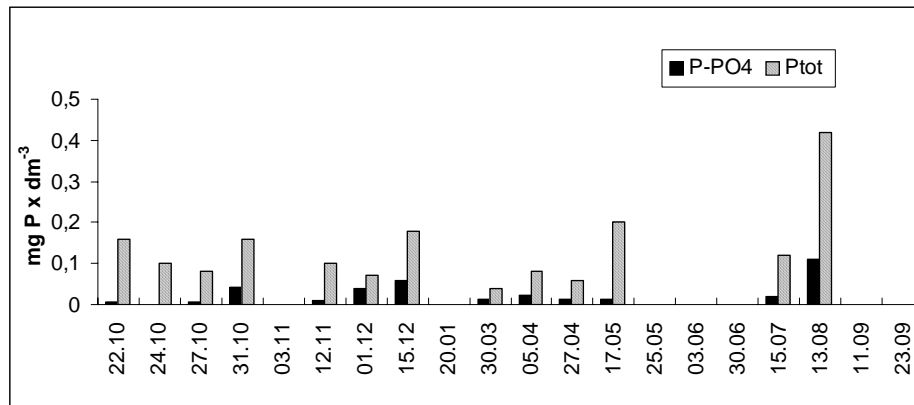


Fig. 3. Influence of the number of trichomes on the emergence of akinetes.

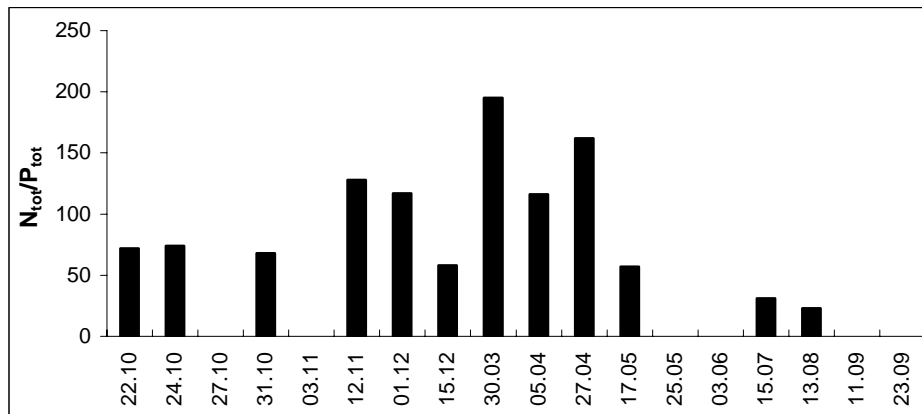
The akinetes of *A. flos-aquae* are cylindrical, wider than vegetative cells ( $7.2-8.0 \mu\text{m}$  wide and  $41.5-56.0 \mu\text{m}$  long) and set in the trichomes with no relation to the heterocysts (Fig. 1c). At the end of October and the beginning of

November, a substantial brittleness of trichomes was noticed, possibly caused by the arrival of ice cover and a deterioration of trophic conditions. The appearance of akinetes was first recorded in the time of the largest occurrence of trichomes, and their sudden increase in quantity at the end of population growth period (Fig. 3). In that period the amount of free akinetes in a milliliter ranged between 628 and 12,300.

Figure 4 shows the concentration of orthophosphates and total phosphorus. During the research, low concentrations of this nutrient were detected. The average amount of P-PO<sub>4</sub> was 0.03 mg x dm<sup>-3</sup> and P<sub>tot</sub> 0.14 mg x dm<sup>-3</sup>. A high N<sub>tot</sub>: P<sub>tot</sub> ratio continued throughout the research period (Fig. 5).



**Fig. 4.** Changes in the concentration of orthophosphates and total phosphorus over the period of research.



**Fig. 5.** Changes in the ratio of total nitrogen to total phosphorus over the period of research.

## DISCUSSION

Comparing the concentration of two basic nutrients - N and P – in the artificial water body shows a clear domination of nitrate nitrogen, which actually is the only form of mineral nitrogen. A similarly high concentration of mineral nitrogen, accompanied by the bloom of *Aphanizomenon flos-aquae*, was also observed in the lakes near Gniezno, however the dominating nitrate form was ammonia nitrogen there (Burchardt 1987, Burchardt & Pańczakowa 1987, Kokociński *et al.* 2002). In the subject pond the concentration of N-NH<sub>4</sub> fluctuated between 0.05 and 0.37 mg x dm<sup>-3</sup>. A good oxygenation of water (11-18 mg O<sub>2</sub> x dm<sup>-3</sup>) provides favourable conditions for mineralization, resulting in formation of nitrates. Large amounts of total nitrogen are likely to be connected with the considerable amount of organic matter supplying the water body in the form of fallen leaves and bird excrement. The concentration of the other nutrient, *i.e.* phosphorus, is quite low (Fig. 4). On the other hand, the amount is not low enough to assume for this element an inhibiting role for the growth of algae. According to Takano and Hino (2000), in low-phosphorus conditions algae can still develop, using the nutrients that have been internally accumulated (so-called internal nutrients) and then also the nutrients released by zooplankton and dead algae (so-called recycled nutrients). Thus, the biomass of *Aphanizomenon flos-aquae* may increase 8 or 12 times without external supply, provided the initial accumulation of P is sufficient. *A. flos-aquae* reveals faster adaptation to the conditions of limited concentration of phosphorus at 15°C than when the temperature ranges from 20 to 25 °C .

According to Barica (1994) and Dokulil & Teubner (2000), the factor which triggers the domination of blue-green algae is the low N<sub>tot</sub>:P<sub>tot</sub> ratio. However, in the subject pond the average ratio at which the growth of *Aphanizomenon flos-aquae* was observed was 67, therefore in this particular water body the nutrients ratio (N<sub>tot</sub>:P<sub>tot</sub>) had no impact on the bloom (Fig. 5).

*Aphanizomenon flos-aquae* is found in the phytoplankton of the majority of medium lakes in a vast range of temperatures. It develops well both at 25°C and when the temperature falls to 10°C (Anagnostidis & Economou-Amilli 1988, Tsujimura *et al.* 2001). Even the emergence of a thin ice cover on 24 October did not inhibit the growth of the blue-green algae (Fig. 3). However, at that time *A. flos-aquae* did not produce the characteristic bunches on the water surface any longer. Jones (1979) associates the formation of heterocysts in a trichome with the presence of nitrates in the water. An increase of N-NO<sub>3</sub> concentration in water results in a decrease of the amount of heterocysts, while a decrease in nitrates brings an increase in those cells. A similar correlation could be observed in the subject pond (Fig. 2).

The identification of the factors controlling the emergence of akinetes in a natural environment is quite complex and difficult to recognize. Perhaps their development is a form of response to one of many unfavourable environmental conditions (Jones 1979, Anagnostidis & Economou-Amilli 1988). An accelerated growth of akinetes was observed at the time of gradual decrease of the amount of *Aphanizomenon flos-aquae* (Fig. 3) caused by a deterioration of its habitat. Rother & Fay (1977, cit. by Jones 1979) noticed the occurrence of akinetes at the time of large densification of population. In the subject pond, the akinetes first emerged at the moment of the largest density of *A. flos-aquae* (90,785 trichomes in a milliliter). The subsequent growth of blue-green algae was insignificant and no akinetes were observed. Jones (1979) does not rule out the influence of a high reaction of the water (9.33-9.61) to the presence of akinetes, however in the subject water body the pH kept quite low (7.4-8.4).

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