

## NUMBER OF NUCLEI, MITOTIC ACTIVITY AND CELL LENGTH IN *CLADOPHORA SP* THALLUS TREATED WITH CADMIUM AND CHROMIUM

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### ABSTRACT

*Cladophora sp.*, a fresh water, filamentous, multi-nucleate alga growing 16 days in the presence of cadmium and chromium at concentrations  $10^{-4}$ - $10^{-8}$ M was the subject of the experiment. Chromium ions reduced the number of nuclei and mitotic activity, and disturbed the correlation between cell length and number of nuclei, more than cadmium ions. Moreover, both tested metals caused the disappearance of cells with numerous nuclei with time of the culture. Only during the first (1-4) days of culture for both metals the concentration of  $10^{-4}$ M and especially of  $10^{-8}$ M increased the number of nuclei, mitotic index and the length of cells. Apical cells were more sensitive to metals than other thallus cells.

KEY WORDS: metals, number of nuclei, mitotic activity, cell length, *Cladophora sp.*

### INTRODUCTION

Metals introduced to the atmosphere by man are uptaken by algae which are important primary producers of organic matter. In algae treated with cadmium and chromium disturbances in mobility (Bassi and Donini 1984; Rachlin et al. 1984) and in the number and shape of cells (Fasulo et al., 1982, 1983) were observed. Moreover, changes in nuclear DNA content (Bonaly et al. 1980) and abnormalities in cell ultrastructure (Heumann 1987) were also reported.

Most of studies concerned mono-nucleate microorganisms (Fasulo et al. 1982, 1983; Rachlin et al. 1982 *a,b,c*, 1983). No data referring to metal influence on multi-nucleate algae are available. Mechanisms of toxicity are also scarcely known in these plants. Therefore, the present experiments were conducted on filamentous, multi-nucleate alga *Cladophora sp.* The number of nuclei, mitotic activity and cell length were analysed in prolonged metal presence.

Chromium necessary for normal development and function of plants and cadmium (heavy metal) whose role has not been specified yet, were used. We tried to establish the metals concentration at which toxic effects were easily observed.

### MATERIAL AND METHODS

Multi-nucleate cells of filamentous alga *Cladophora sp.* (*Cladophoraceae*) from fresh water pond in Rogów arboretum were the object of the present studies. Algae freed from organic remnants, were put into water solutions of cadmium  $\text{Cd}(\text{NO}_3)_2 \times 4\text{H}_2\text{O}$  or chromium  $\text{Cr}(\text{NO}_3)_3 \times 9\text{H}_2\text{O}$  in concentra-

tions from  $10^{-4}$  to  $10^{-8}$ M prepared on pond water. Control material was cultivated on Rogów pond water. *Cladophora sp.* was grown for 16 days at room temperature (18-22°C) in normal photoperiod. To maintain the concentration of tested metals on the constant level the solutions were changed every for 4 days by supplementing them with 1/4 of culture medium volume. Before and after the experiment pH of media were measured. The culture solutions were aerated three hours every day.

After 1, 4, 8 and 16 days of culture the material was fixed in Carnoy (ethanol, acetic acid, 3:1) for 24 hrs at 4°C. Following several washings in 70% alcohol the fragments of thallus were stained with hot acetocarmine. The number of nuclei, mitotic index and cell length were analysed in 30 apical and 130 other thallus cells.

### RESULTS

In apical cells growing in natural pond water the number of nuclei decreased with time reaching minimum on the 8th day of culture (Table 1). On the last (16th) day the number of nuclei was similar as at the beginning (1st day) of the experiment. Similar results were observed in the material treated with cadmium at the concentration of  $10^{-7}$ M. However, no significant differences in the number of nuclei were observed in the presence of chromium with the exception of  $10^{-8}$ M concentration at which the number of nuclei decreased only on the 16th day of the experiment.

The addition of the metals to pond water affected the number of nuclei in apical cells in various ways (Table 1). The

TABLE 1. Number of nuclei in apical cells of *Cladophora sp.*

| Material<br>metal (M) | TIME OF CULTURE (DAYS) |          |          |          |          |          |          |          |          |          |          |          |
|-----------------------|------------------------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
|                       | 1                      |          |          | 4        |          |          | 8        |          |          | 16       |          |          |
|                       | 0                      | Cd       | Cr       | 0        | Cd       | Cr       | 0        | Cd       | Cr       | 0        | Cd       | Cr       |
| 0                     | 21.2±1.3               | –        | –        | 15.7±1.2 | –        | –        | 11.5±0.9 | –        | –        | 18.7±1.7 | –        | –        |
| 10 <sup>-4</sup>      | –                      | 15.3±1.7 | 14.3±0.9 | –        | 15.5±1.6 | 14.0±0.7 | –        | 12.9±1.0 | 14.7±0.8 | –        | 8.9±0.6  | 13.4±1.2 |
| 10 <sup>-5</sup>      | –                      | 18.7±1.1 | 13.2±1.3 | –        | 16.4±1.0 | 16.7±0.9 | –        | 13.8±1.1 | 13.6±0.8 | –        | 12.0±0.7 | 12.7±0.8 |
| 10 <sup>-6</sup>      | –                      | 21.7±2.0 | 17.3±0.8 | –        | 14.4±1.3 | 16.7±0.9 | –        | 18.0±1.5 | 17.7±1.1 | –        | 14.9±1.3 | 17.9±1.2 |
| 10 <sup>-7</sup>      | –                      | 20.9±2.4 | 16.7±1.8 | –        | 16.3±1.2 | 16.5±1.0 | –        | 10.5±1.1 | 16.6±0.8 | –        | 11.9±1.0 | 17.3±1.2 |
| 10 <sup>-8</sup>      | –                      | 32.5±2.0 | 16.2±1.0 | –        | 16.0±0.8 | 15.5±1.0 | –        | 17.4±1.4 | 17.2±1.0 | –        | 11.7±1.3 | 12.7±0.6 |

TABLE 2. Number of nuclei in other thallus cells of *Cladophora sp.*

| Material<br>metal (M) | TIME OF CULTURE (DAYS) |          |          |          |          |          |         |          |          |          |          |          |
|-----------------------|------------------------|----------|----------|----------|----------|----------|---------|----------|----------|----------|----------|----------|
|                       | 1                      |          |          | 4        |          |          | 8       |          |          | 16       |          |          |
|                       | 0                      | Cd       | Cr       | 0        | Cd       | Cr       | 0       | Cd       | Cr       | 0        | Cd       | Cr       |
| 0                     | 15.5±0.7               | –        | –        | 10.8±0.3 | –        | –        | 9.7±0.3 | –        | –        | 13.3±0.4 | –        | –        |
| 10 <sup>-4</sup>      | –                      | 11.5±0.5 | 10.6±0.9 | –        | 11.2±0.4 | 10.0±0.8 | –       | 12.7±0.5 | 8.3±0.7  | –        | 11.2±0.5 | 9.2±0.8  |
| 10 <sup>-5</sup>      | –                      | 12.5±0.5 | 6.3±1.0  | –        | 13.9±0.5 | 7.0±0.6  | –       | 11.1±0.4 | 6.4±0.5  | –        | 11.0±0.4 | 6.2±0.5  |
| 10 <sup>-6</sup>      | –                      | 15.3±0.6 | 13.3±1.1 | –        | 10.6±0.5 | 6.4±0.6  | –       | 14.7±0.6 | 9.9±0.8  | –        | 10.0±0.5 | 10.4±0.8 |
| 10 <sup>-7</sup>      | –                      | 13.4±0.6 | 11.1±0.9 | –        | 12.6±0.4 | 7.6±0.7  | –       | 9.8±0.4  | 12.0±1.0 | –        | 10.2±0.5 | 6.8±0.6  |
| 10 <sup>-8</sup>      | –                      | 19.0±0.7 | 13.0±1.1 | –        | 9.9±0.4  | 6.2±0.5  | –       | 13.5±0.6 | 8.3±0.7  | –        | 9.2±0.3  | 7.1±0.6  |

number of nuclei underwent reduction in all cadmium concentrations on the 16th day and in 10<sup>-4</sup>M on the 1st day. In the presence of chromium in tested concentrations the diminution in number of nuclei in apical cells was seen on the 1st day and at the concentrations of 10<sup>-4</sup>-10<sup>-5</sup>M on the 16th day of culture. On the other hand, treatment with the tested metals increased the number of nuclei in apical cells. It was observed in the presence of cadmium at concentration 10<sup>-8</sup>M on the 1st and 8th days and at 10<sup>-6</sup>M on the 8th day. Higher number of nuclei in apical cells treated with chromium at concentrations from 10<sup>-6</sup> to 10<sup>-8</sup>M were seen on the 8th day. In control plants the number of nuclei in other thallus cells similarly as in apical cells decreased with time of culture reaching minimum on the 8th day (Table 2). On the last day of the experiment the number of nuclei was similar as on the 1st day. Similar tendency, with the exception of the last (16th) day, was observed in the presence of cadmium at the concentration 10<sup>-7</sup>M.

The addition of cadmium and mostly chromium to pond water changed the number of nuclei in other thallus cells of *Cladophora sp.* Decrease in the number of nuclei was seen in the presence of chromium at the concentration 10<sup>-4</sup>M on the 1st and 16th days while in the presence of cadmium at the concentrations from 10<sup>-6</sup> to 10<sup>-8</sup> M on the 16th day (Table 2). The exception were chromium treated algae at the concentration 10<sup>-4</sup>M on the 4th and 10<sup>-8</sup>M on the 8th day where no significant changes in the number of nuclei appeared. Metals presence in pond water increased also the number of nuclei in other than apical *Cladophora sp.* thallus cells (Table 2). This effect was observed in the presence of cadmium at the concentration 10<sup>-8</sup>M on the 1st day, at the concentrations 10<sup>-5</sup> and 10<sup>-7</sup>M on the 4th day and at the concentrations 10<sup>-4</sup>, 10<sup>-6</sup> and 10<sup>-8</sup>M on the 8th day of culture. A slight decrease in the num-

ber of nuclei was also noticed in the presence of chromium at the concentration 10<sup>-7</sup>M on the 8th day of culture.

Due to significant differences in the number of nuclei as well as in pH of culture media at the beginning and at the end of the experiment (Table 3) cadmium and chromium at the concentrations 10<sup>-4</sup> and 10<sup>-8</sup>M were used in further studies.

In control material on the 1st day of experiment apical cells with 8 to 32 nuclei were observed but cells with mean number of nuclei (12-20) dominated (Fig. 1). On the 4th and especially 8th day the number of cells with 21-32 nuclei decreased while those with 4-16 nuclei increased. On the last day of experiment cells with 14 nuclei dominated and multinucleate cells with 21-32 nuclei reappeared.

The addition of chromium and cadmium at both studied concentrations (10<sup>-4</sup>M, 10<sup>-8</sup>M) to pond water changed the

TABLE 3. Changes in pH of medium during 16 days of culture

| Material<br>Metal (M) | TIME OF CULTURE (DAYS) |     |     |     |     |     |
|-----------------------|------------------------|-----|-----|-----|-----|-----|
|                       | 1                      |     |     | 16  |     |     |
|                       | O                      | Cd  | Cr  | O   | Cd  | Cr  |
| O                     | 8                      | –   | –   | 8.3 | –   | –   |
| 10 <sup>-4</sup>      | –                      | 7.9 | 7.7 | –   | 8.1 | 7.6 |
| 10 <sup>-5</sup>      | –                      | 8.1 | 8.0 | –   | 8.2 | 8.3 |
| 10 <sup>-6</sup>      | –                      | 8.2 | 8.1 | –   | 8.2 | 8.4 |
| 10 <sup>-7</sup>      | –                      | 8.2 | 8.1 | –   | 8.3 | 8.4 |
| 10 <sup>-8</sup>      | –                      | 8.2 | 8.1 | –   | 8.3 | 8.4 |

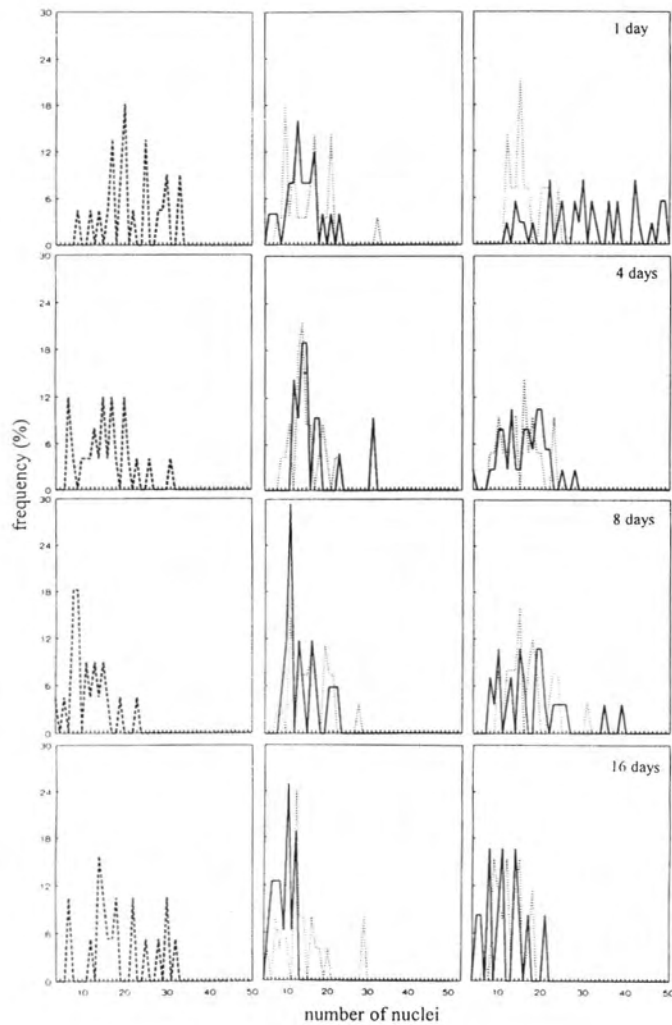


Fig. 1. Frequency of appearance of apical cells with different number of nuclei during 16-day a culture in the presence of cadmium (—) and chromium (...) ions at concentrations  $10^{-4}$  (middle) and  $10^{-8}$  M (right); control (---)

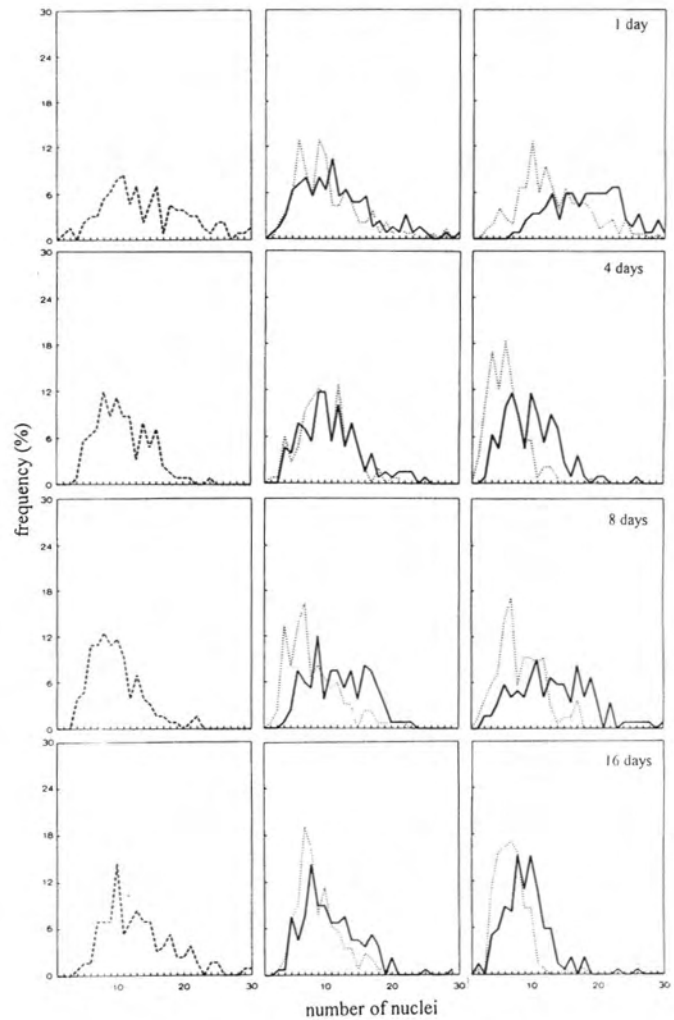


Fig. 2. Frequency of appearance of other thallus cells with different number of nuclei during a 16-day culture in the presence of cadmium (—) and chromium (...) ions at concentrations  $10^{-4}$  (middle) and  $10^{-8}$  M (right); control(---).

number of multi-nucleate apical cells (Fig. 1). On the 1st day in the presence of both metals at the higher concentration cells with 2-21 nuclei were present while in material treated with chromium and cadmium at  $10^{-8}$  M nuclei with 12-24 and 12-48 cells, respectively were observed. With time of culture in metals presence at both concentrations the cells with many nuclei (21-28) were less frequent, it was especially visible in the presence of cadmium at the concentration  $10^{-4}$  M on the 16th day.

Other thallus cells of control plants contained a varied number of nuclei (2-32) within a single cell (Fig. 2). On the 4th and 8th days of culture cells with a high number (24-32) of nuclei disappeared but some such cells reappeared on the last (16th) day of the experiment. Similar tendency was observed in the presence of cadmium and chromium at the concentrations  $10^{-4}$  and  $10^{-8}$  M (Fig. 2). The exception was material treated with cadmium at the concentration  $10^{-8}$  M, where on the 1st day no cells with small number of nuclei were observed and on the 8th day small number of multi-nucleate cells (22-30) appeared with time. In the presence of both metals the number of cells with high number of nuclei decreased which was especially visible after chromium treatment.

In *Cladophora sp.* cells the most common mitotic figures were prophase, later stages of mitosis appeared rarely and therefore were omitted. In control material prophase index first increased reaching maximum on the 4th day (Table 4, 5) then decreased rapidly. On the 16th day the prophase were not visible in apical cells.

Complete reduction of mitotic activity was characteristic for thallus of *Cladophora* growing in laboratory for more than three weeks. The next wave of mitoses appeared usually 6-8 weeks later. Although we tried to establish the factors triggering the new wave of mitoses we still do not know what stimulates the nuclei to division. Our observations allow us to suppose only that light and/or temperature play an important role in this process.

Similar, as in control material, pattern of changes in mitotic activity was observed in all thallus cells in the presence of cadmium at the concentration  $10^{-4}$  M and partly in other cells of thallus treated with chromium at the concentration  $10^{-8}$  M (Table 4, 5).

Mitotic activity disappeared on the 8th day in apical cells in the presence of chromium at the concentration  $10^{-4}$  M while in other thallus cells this effect was seen on the same day but in

TABLE 4. Mitotic activity in apical cells of *Cladophora sp.* (%)

| Material<br>metal (M) | TIME OF CULTURE (DAYS) |      |     |      |      |     |     |     |     |     |     |     |
|-----------------------|------------------------|------|-----|------|------|-----|-----|-----|-----|-----|-----|-----|
|                       | 1                      |      |     | 4    |      |     | 8   |     |     | 16  |     |     |
|                       | 0                      | Cd   | Cr  | 0    | Cd   | Cr  | 0   | Cd  | Cr  | 0   | Cd  | Cr  |
| 0                     | 14.1                   | –    | –   | 28.7 | –    | –   | 0.8 | –   | –   | 0.0 | –   | –   |
| $10^{-4}$             | –                      | 12.7 | 4.4 | –    | 40.8 | 2.7 | –   | 1.3 | 0.0 | –   | 2.7 | 3.6 |
| $10^{-8}$             | –                      | 17.8 | 1.3 | –    | 0.0  | 2.8 | –   | 0.0 | 1.4 | –   | 1.4 | 0.0 |

TABLE 5. Mitotic activity in other thallus cells of *Cladophora sp.* (%)

| Material<br>metal (M) | TIME OF CULTURE (DAYS) |      |     |      |      |     |     |     |     |     |     |     |
|-----------------------|------------------------|------|-----|------|------|-----|-----|-----|-----|-----|-----|-----|
|                       | 1                      |      |     | 4    |      |     | 8   |     |     | 16  |     |     |
|                       | 0                      | Cd   | Cr  | 0    | Cd   | Cr  | 0   | Cd  | Cr  | 0   | Cd  | Cr  |
| 0                     | 12.3                   | –    | –   | 26.9 | –    | –   | 1.9 | –   | –   | 0.5 | –   | –   |
| $10^{-4}$             | –                      | 15.2 | 3.9 | –    | 45.7 | 6.6 | –   | 2.8 | 2.4 | –   | 0.0 | 2.0 |
| $10^{-8}$             | –                      | 26.7 | 1.7 | –    | 1.3  | 4.4 | –   | 1.1 | 0.0 | –   | 0.0 | 0.0 |

that metal presence at the concentration  $10^{-8}$ M. Cadmium treatment ( $10^{-8}$ M) caused a lack of mitoses in apical cells on the 4th day but in both concentrations used on the 16th day of culture in other cells of thallus.

Both tested metals presence in culture medium affected mitotic activity in *Cladophora sp.* thallus cells in various ways. In apical cells cadmium at the concentration  $10^{-4}$ M increased prophase index on the 4th-16th days and at  $10^{-8}$ M on the 1st and 16th days; while in the presence of chromium ( $10^{-4}$ M) the increased number of prophases was seen only on the 16th day (Table 4).

In other thallus cells similar effects of metal action appeared at both metals concentrations on the 1st day and at concentration  $10^{-4}$ M on the 4th and 8th days as well. The increase in the number of prophases after chromium treatment at the concentration  $10^{-4}$ M was observed on the 8th and 16th days of experiment (Table 5).

Besides stimulation the tested metals reduced the number of prophases in all *Cladophora sp.* cells (Table 4, 5). Lower

number of prophases was observed in the presence of chromium at both concentrations ( $10^{-4}$ ,  $10^{-8}$ M) on the 4th and 8th days. Moreover, in apical cells diminution in number of prophases took place on the 1st day in the presence of cadmium at the concentration  $10^{-4}$ M and in other cells of thallus on the 16th day at the concentration  $10^{-8}$ M.

While the length of apical cells in control material did not change during experiment (Fig. 3) that of other ones significantly increased with time reaching maximum on the 16th day (Fig. 4). Similar pattern of cell growth was observed in the presence of both metals at the low concentration ( $10^{-8}$ M) and slight growth on the 4th and 8th days in the presence of chromium at the concentration  $10^{-4}$ M. (Figs 3, 4). Only in the material treated with both metals but mostly cadmium at the concentration  $10^{-4}$ M the effect was reversed, i.e. decrease in the length of cells with time of culture.

Addition of both tested metals at the concentration  $10^{-4}$ M to pond water decreased the length of thallus cells while at  $10^{-8}$ M it increased it especially on 8th and 16th days of the experiment

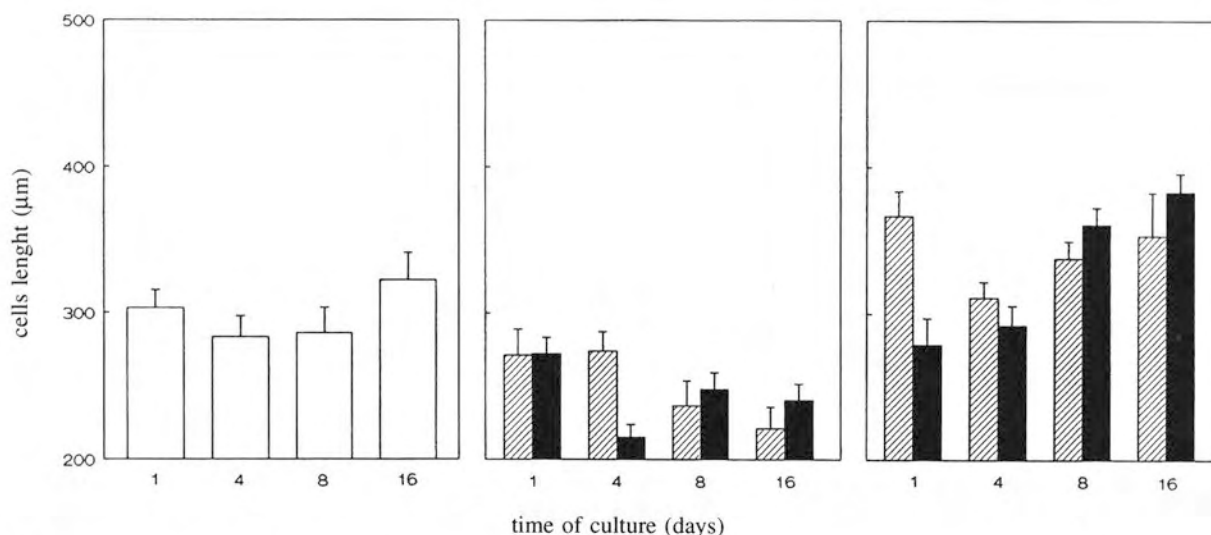


Fig. 3. Relation between length of apical cells and number of nuclei during a 16-day culture in the presence of cadmium (striped field) and chromium (black field) at concentrations  $10^{-4}$  (middle) and  $10^{-8}$ M (right); control (white).



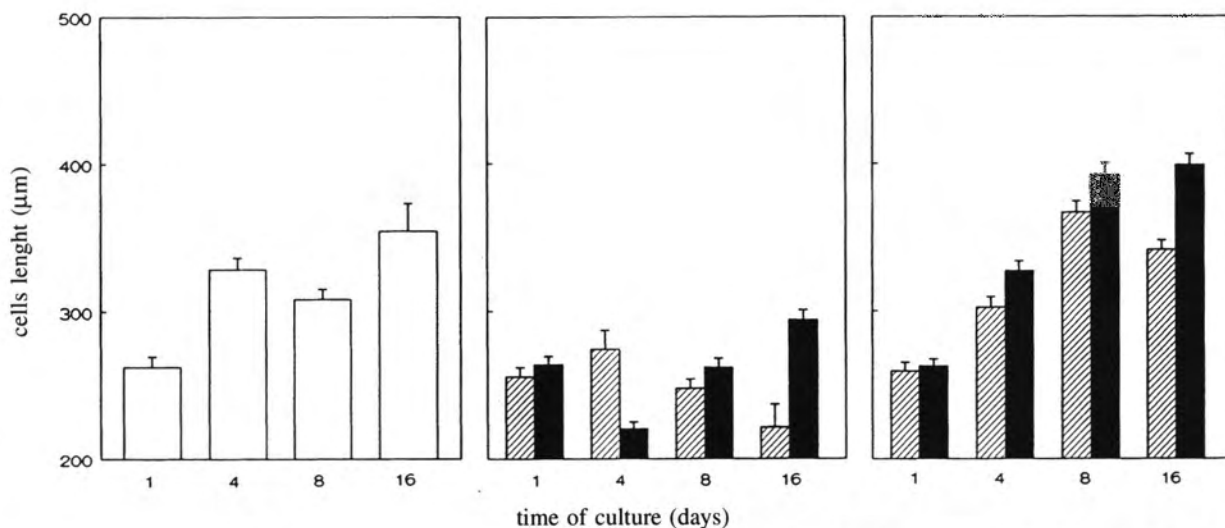


Fig. 4. Relation between length of other thallus cells and number of nuclei during 16-day culture in the presence of cadmium (striped field) and chromium (black field) at concentrations  $10^{-4}$  (middle) and  $10^{-8}$  M (right); control (white).

(Figs 3, 4). The length of *Cladophora sp.* cells was positively correlated with the number of nuclei (Figs 5, 6). No such relation was observed in a small group of cells with various number of nuclei during the whole experiment. Cadmium and chromium ions at both concentrations disturbed that correlation that was mostly visible in apical cells (Fig. 5) treated with chromium at both concentrations during the whole experiment and with cadmium at the concentration  $10^{-4}$  M from the 1st till 8th day. In other thallus cells (Fig. 6) no correlation between length of cells and the number of nuclei was noticed in chromium presence at both concentrations on the 1st day and at  $10^{-4}$  M till the 8th day of culture. The same effect was observed in the presence of cadmium at the concentration  $10^{-4}$  M on the 4th and 16th days of culture.

## DISCUSSION

The data obtained indicate that the tested metals presence in pond water decreased mitotic activity of *Cladophora sp.* thallus cells which brought about diminution in the number of nuclei and consequently in the amount of multi-nucleate cells. Reduced mitotic activity was also observed in uni-cell alga *Euglena gracilis* treated with cadmium (Bonaly et al. 1980) and chromium (Fasulo et al. 1982, 1983). Similar effects were reported in roots of flower plants growing in the presence of heavy metals (Wierzbicka 1989; Gabara and Gołaszewska 1993; Zhang and Yaung 1994). Heavy metals form stable complexes with nucleic acids and disturb DNA replication (Bianchi et al. 1977; Levis et al. 1978; Bonaly et al. 1980).

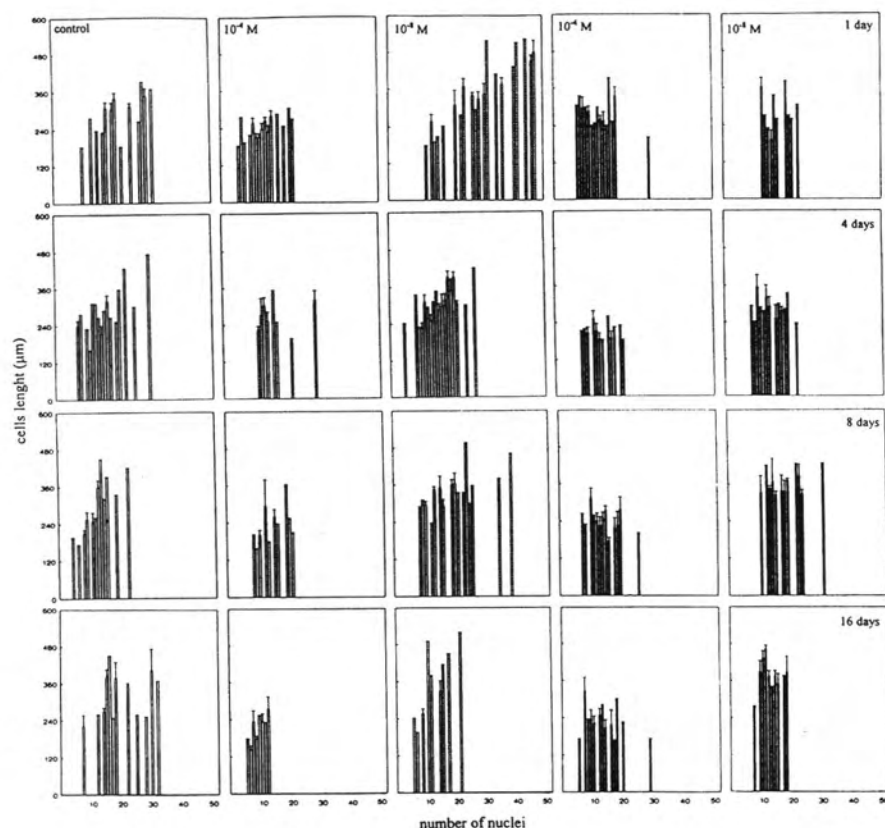


Fig. 5. Mean length of apical cells growing in a 16-day culture in the presence of cadmium (striped field) and chromium (black field) at concentrations  $10^{-4}$  and  $10^{-8}$  M.

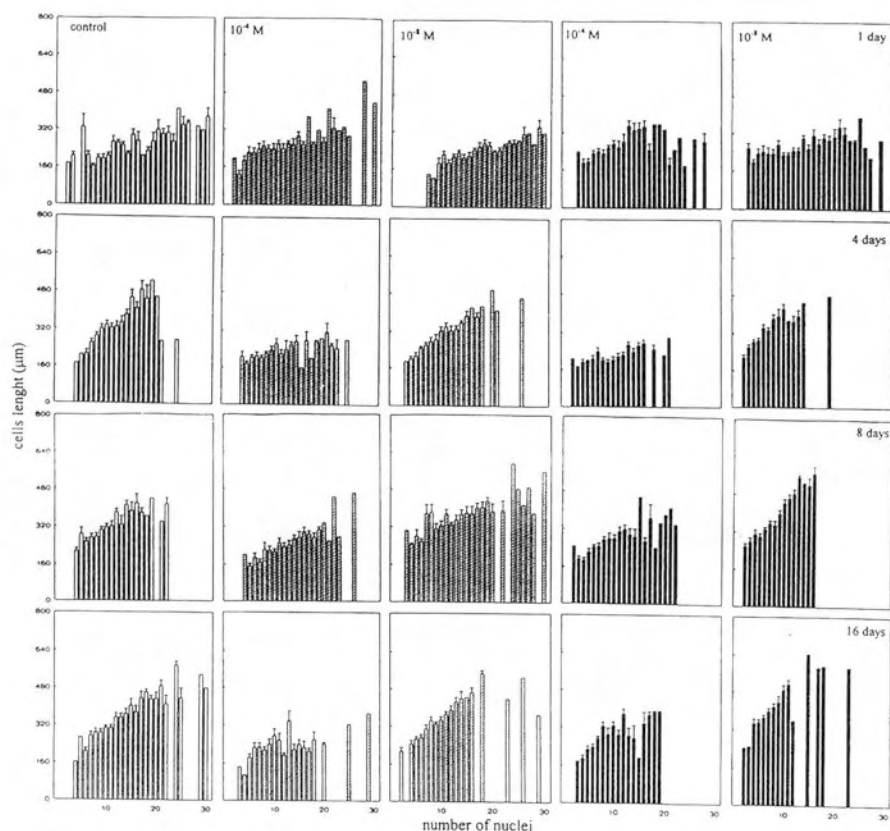


Fig. 6. Mean length of other thallus cells growing in a 16-day culture in the presence of cadmium (striped field) and chromium (black field) at concentrations  $10^{-4}$  and  $10^{-8}$ M.

According to Stobardt et al. (1985) opinion disturbances in DNA replication are caused by metals binding with SH groups and in consequence blocking DNA polymerase activity. Lower radioactive thymidine incorporation into nuclei of meristematic cells of pea root (Gabara et al. 1992), increase in the number of 2C DNA cells with simultaneous decrease in the number of 4C DNA cells after heavy metal treatment (Wojtyła-Kuchta and Gabara 1991, Stecka et al. 1995) seem to support the hypothesis of heavy metals influence on DNA replication.

Metals seem to act specifically in G<sub>2</sub> phase or in early stages of mitosis thus blocking cells in pre-mitotic phase (Fasulo et al. 1983, Corradi et al. 1991). Consequently, lower mitotic activity and smaller number of nuclei in *Cladophora sp.* cells might result from the lack of proteins necessary for progression of cells from G<sub>2</sub> to M phase.

Increase in the number of nuclei accompanied by higher mitotic activity in *Cladophora sp.* cell mostly in the presence of cadmium at the concentration  $10^{-4}$ M on the 4th day of culture, is difficult to explain in reference to the above data. The only case known to the author of increase in the number of nuclei was reported in *Euglena gracilis* treated with Cr<sup>6+</sup> (Fasulo et al. 1982, 1983). However, the higher number of nuclei in this alga was caused by fusion of mono-nucleate cells. In Melnishuk et al. (1982) opinion short-time treatment with heavy metals not only stimulated transcription and translation but increased DNA replication as well. Thus, one can assume that increase in prophase index and in the number of nuclei in *Cladophora sp.* cells could be the consequence of these processes.

According to Wik-Sjöstedt (1970) mitoses in salt-water *Cladophora sp.* took place in the morning hours in a day-night cycle, while in fresh water species being the object of the present studies they occur in the evening (unpubl.). Changes in the number of nuclei and mitotic activity in the

presence of the tested metals could result from disturbances in *Cladophora* cell cycle. Prolongation of cell cycle to 25 or even 40 hours observed in root meristem growing in the presence of heavy metals (Wierzbicka, 1992) might support the above hypothesis.

The seasonal growth of *Cladophora* thallus (spring-autumn) is especially intensive in spring (Whitton et al. 1981). The growth of thallus is caused by divisions and elongation of mainly apical cells. The results obtained in the present experiment show that these cells are more sensitive to the tested metals than other thallus cells.

*Cladophora sp.* is one of the species in which cytokinesis does not result directly from nuclear divisions (Gabara 1965, Mc Donald and Pickett-Heaps 1976) although the increase in the number of nuclei seems to be one of the factors triggering cytokinesis (Gabara 1965). The results of the present paper clearly demonstrate the existence of correlation between the length of cells and the number of nuclei in *Cladophora sp.* This regularity was disturbed especially in apical cells growing in the presence of both tested metals at the concentration  $10^{-4}$ M. Elongation of apical and other thallus cells was also disturbed in *Cladophora sp.* Inhibitory effect of metals on cell length was also observed in other mono-nucleate algae *Plectonema boryanum* (Rachlin et al. 1982 c) *Chlorella saccharopilia* (Rachlin et al. 1982a, b) *Navicula incerta* (Rachlin et al. 1982 b, 1983) *Anabaena flos-aquae* (Rachlin et al. 1984) *Nitzschia closterium* (Rachlin et al. 1982 b), and multi-nucleate algae *Chara vulgaris* (Heumann 1987). Decrease in the cell length in the presence of heavy metals might be, according to De Fillipis et al. (1981) the effect of NADP-oxydoreductase inhibition since metals have high affinity to sulphhydryl groups of this enzyme. Małkowski (1993) reported that in coleoptyles of crops lead ions disturbed the activity of plasmalemma ATPase. Abnormal permeability of membranes especially of plasmalemma might be responsible for limited

elongation of cells. Inhibition of elongation could also be due to changes in wall resistance caused by substitution of calcium ions in pectin substances with heavy metal ions (cf. after Małkowski 1993).

The data presented in this paper show stimulated elongation of *Cladophora* cells in the presence of both metals at the concentration  $10^{-8}$ M. Enlargement of cells is one of the commonly observed effects of heavy metals on both uni-celled organisms (Mertz 1969, Fasulo et al. 1982, 1983) and multi-celled ones (Gabara unpubl.). This growth may be the result of enhanced vacuolization caused by disturbances in membrane permeability (Fasulo et al. 1983) because metals showing affinity to SH groups (Fasulo et al. 1983) change physico-chemical features of membranes (Vallee and Ulmer 1972), especially composition of lipids (Ros et al. 1990).

At present it is difficult to explain higher toxicity of chromium, a microelement, than of cadmium (heavy metal) whose negative effects are widely described in literature (Bonaly et al. 1980; De Fillipis et al. 1981; Rachlin et al. 1982a, b, c, 1983; Heumann 1987; Wojtyła-Kuchta and Gabara 1991; Gabara et al. 1992; Gabara and Gołaszewska 1993). Changes in pH of pond water containing chromium ions at the beginning and in the end of the experiment seem to be partly responsible for increased toxicity of this metal.

In conclusion, due to significant effects of cadmium and chromium at the concentrations  $10^{-4}$  and  $10^{-8}$ M on the 4th and especially on the 8th day of culture in further experiments with metals effects on *Cladophora* cells 16-day culture will be omitted.

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LICZBA JĄDER, AKTYWNOŚĆ MITOTYCZNA ORAZ DŁUGOŚĆ KOMÓREK PLECHY  
*CLADOPHORA SP.* TRAKTOWANYCH KADMEM I CHROMEM

## STRESZCZENIE

Obiektem badań był słodkowodny, nitkowaty, wielojądrowy glon *Cladophora sp.* poddany 16-dniowemu działaniu dwóch metali: kadmu i chromu w stężeniu  $10^{-4}$ - $10^{-8}$ M. Jony chromu w stężeniu  $10^{-4}$ M i  $10^{-8}$ M w większym stopniu, aniżeli jony kadmowe, redukowały liczbę jąder, aktywność mitotyczną oraz zakłócały korelację między długością komórki a liczbą jąder. W miarę trwania hodowli oba testowane metale powodowały ponadto zanik populacji komórek o dużej liczbie jąder. Jedynie w początkowych (1-4) dniach hodowli dodatek testowanych metali w stężeniu  $10^{-4}$ M a zwłaszcza  $10^{-8}$ M zwiększał liczbę jąder, wartość indeksu mitotycznego oraz długość komórek. Komórki szczytowe okazały się bardziej wrażliwe na testowane metale aniżeli pozostałe komórki plechy.

SŁOWA KLUCZOWE: metale, liczba jąder, aktywność mitotyczna, długość komórek, *Cladophora sp.*