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Morphological Changes in the Diatom, *Tabellaria flocculosa*, Induced by Very Low Concentrations of Cadmium

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As is well known, algae are greatly affected by toxic metals in natural waters (LAUBE et al. 1980, SYMPOSIUM 1978). Besides inhibiting growth, such metals may also affect cell morphology. Copper can cause giant cell formation in *Chlorella vulgaris* (FOSTER 1977) and *Ankistrodesmus braunii* (MASSALSKI et al. 1981). Cu or Cd distorted or lysed cells of *Anabaena* 7120 (MASSALSKI et al. 1981). Mercury can cause giant cell formation in a *Chlorella* sp. and the green flagellate *Dunaliella tertiolecta* (DAVIES 1976). MONAHAN (1976a) found that though *Scenedesmus obtusiusculus* normally grows as colonies, in the presence of 50 µg/L Cd it grew mainly as single cells. Toxic Pb concentrations caused similar morphological changes in this alga (MONAHAN 1976b). TOMPKINS & BLINN (1976) found that sublethal concentrations of Hg caused a striking morphological change in the planktonic diatom *Asterionella formosa*. Instead of forming their usual stellate colonies of 8-16 cells, cultures formed large cylindrical stacks composed of 25-30 colonies.

The diatom, *Tabellaria flocculosa*, is widely distributed in the lakes and streams of North America. We describe here a morphological change in this organism which is specifically induced by very low levels of Cd.

EXPERIMENTAL

The alga, *T. flocculosa*, was isolated from Lake Carriere, Point Gatineau, Québec and identified from the description given by KOPPEN (1975). The growth medium was based on Chu's No. 10 medium as modified by GERLOFF et al. (1950). We found that growth of *T. flocculosa* was greatly improved by addition of an extract made by autoclaving a mixture of equal volumes of peat moss (commercial garden grade) and deionized water, cooling and filtering. The medium contained the following components (in mg/L): $Ca(NO_3)_2 \cdot 4H_2O$ (400): K_2HPO_4 (100): $MgSO_4 \cdot 7H_2O$ (25): $Na_2SiO_3 \cdot 9H_2O$ (5): Na_2CO_3 (20): Ferric Citrate (3): Na_2CO_3 (20): Citric acid (3): and Peat-moss extract (10 mL/L): pH 7.2. The medium was sterilized by autoclaving.

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Cultures were grown in 500-mL conical flasks containing 250 mL media, without shaking. Illumination was provided by Westinghouse "cool white" (F40CW) with equal illumination from Westinghouse "agro-lite" (F40/AGRO) fluorescent tubes in a cycle of 16h on, 8h off. The light intensity was $2.4X10^3$ lux. Temperature was maintained at 23°C. Fresh stock cultures were prepared every week by inooculating 50 mL of week-old stock culture into 200 mL fresh medium. Before use glassware was soaked for 30 min in 20% HCl, washed repeatedly with hot water and rinsed several times with glass-distilled water. Metal stock solutions were made up by dissolving the nitrate salt in deionized water and filter sterilizing; aliquots were added to the media in the flasks immediately before innoculation.

RESULTS AND DISCUSSION

Cells of *T. flocculosa* usually form zig-zag chains due to corner-to-corner attachment by gelatinous pads (Fig. 1). The effects of iron, copper, lead, chromium, nickel, zinc and cadmium salts on the growth of *T. flocculosa* are shown in Table 1. At 10^{-7} molar (0.01 mg/L), cadmium inhibited growth more than the other metals. The inhibitory concentration for cadmium is quite clear-cut. Growth was unaffected by 10^{-8} molar (0.001 mg Cd/L). At both 0.01 and 0.001 mg Cd/L, the cells no longer formed the zig-zag configuration; instead they formed the straight-chain configuration shown in Fig. 2. This configuration was not observed at any concentrations of the other metals tested. For example, Fig. 3 shows a culture of *T. flocculosa* grown in 0.001 mg Cu/L ; the zig-zag chain morphology is the same as that of the control culture. This suggests the straight-chain configuration is a unique "cadmium-effect" and not due to a generalised metal toxicity. Concentrations of cadmium lower than 0.001 mg Cd/L did not result in the straight-chain configuration.

The current maximum cadmium level acceptable for drinking water in Canada is 0.02 mg/L; for irrigation, recreational and food processing water supplies the maximum acceptable is 0.01 mg/L (REEDER et al. 1979). We have shown cadmium to be biologically active at 0.001 mg/L, causing the striking morphological changes described above. These are clearly visible in the light microscope.

Due to the extreme sensitivity of *T. flocculosa* for cadmium, we suggest that it might be used as an *in situ* indicator of low level cadmium contamination of water and sediments.

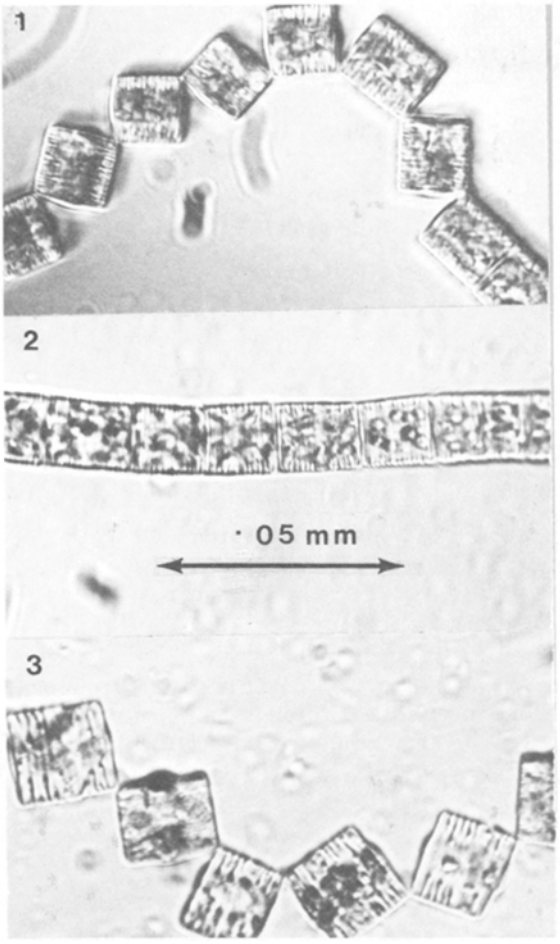

Fig.1. Control culture of *T. flocculosa* grown in absence of toxic metals.

Fig.2. Culture grown in 0.001 mg/L Cd.

Fig. 3. Culture grown in 0.001 mg/L Cu.

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Table 1. Growth of *T. flocculosa* in Different Metals[a]

Metal	Molarity 10^{-5}	10^{-6}	10^{-7}	10^{-8}
Fe	-	+++	+++	++
Cu	-	++	+++	++
Pb	-	+/-	++	++
Cr	-	-	++	++
Ni	-	-	++	++
Zn	-	-	++	++
Cd	-	-	+	++

[a]Visual growth estimates (Approx. Cells/mL)

-	no growth
+/-	slight growth ($\leq 10^3$)
+	moderate growth (2.0×10^4)
++	control growth (3.0×10^4)
+++	more than control (4.0×10^4)

Starting cell concentration was 1×10^3/mL.

Cultures were grown for 14 days at 23°C. Cells were counted by mixing one drop of a 1% rose bengal solution with 1 mL culture, placing 1 µL on a microscope slide and counting the cells directly.

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