CADMIUM UPTAKE AND ITS EFFECT ON THE GROWTH OF CHLORELLA HOMOSPHAERA AND SCENEDESMUS QUADRICAUDA CELLS IN LABORATORY CONDITIONS

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ABSTRACT

The toxic effect and uptake of cadmium during growth of Chlorella homosphaera and Scenedesmus quadrircauda cells was investigated at different initial cell concentrations (0.1 to 2.0 g/l). The cells were grown in synthetic growth medium containing added cadmium chloride to final concentrations ranging from 0.0 to 12.0 mg/l. The growth of Chlorella and Scenedesmus cells was affected by 4.0 mg/l and 2.0 mg/l of the metal, respectively. For both species, this effect was directly proportional to the metallic ion concentration and inversely proportional to the cell concentration.

Key words: cadmium uptake, green microalgae.

INTRODUCTION

The toxic effect of heavy metals on microbial cells is being investigated by various researchers. Metal accumulation promotes several changes in cells, such as alterations in cellular growth (1), cellular division and photosynthetic activity (11). Different microorganisms exhibit mechanisms for the uptake and accumulation of metals, including bacteria (13), fungi (14), marine algae (5, 8), or green microalgae (10).

The interactions microbe-metals are dependent on the source and concentrations of the metallic ion, microbial species involved and physiological and environmental aspects. Previous research indicated that many microorganisms are able to grow in relatively high concentrations of toxic heavy metals by means of different detoxification mechanisms.

This ability is attractive in the recovery of metals (7), or to reduce environmental damage. Several green microalgae, being described in the literature as potential organisms for the uptake of metals, specially in aquatic environments.

Microorganisms from the genus Chlorella and Scenedesmus are unicellular, nutritionally not complex, their sizes ranging from 1.5 to 5.7 µm diameter. Their sources of nitrogen are basically nitrate and urea, being their nitrogen content between 7 to 11%. They are planktonic algae, usually found in freshnesses.

Cells from the genus Chlorella contains autosporas, formed by divisions inside the cells, and chloroplasts showing a narrow opening named "mantel-shaped". Cells from the genus Scenedesmus have their total number of divisions limited by a cell enveloply, previously fixed.

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The aim of the present study was to examine the growth and uptake of cadmium by *Chlorella* *hominophora* and *Scenedesmus quadricauda* at different cadmium ion concentrations, as a study on the employment of these species for the treatment and control of metallurgical effluents.

**MATERIALS AND METHODS**

*Organisms*: Green microalgae *Chlorella hominophora* and *Scenedesmus quadricauda*, isolated from Quinta da Boa Vista Lake (Rio de Janeiro City). The organisms were grown in synthetic medium with the following composition (g/l): NaNO₃, 1.00; KH₂PO₄, 0.25; MgSO₄, 0.50; NH₄Cl, 0.05; CaCl₂, 0.08 and glucose, 10.00. The medium was adjusted to pH 7.0. Culture medium was sterilized at 0.5 atm for 20 minutes.

Experimental procedures: The experiments were conducted in Erlenmeyer flasks containing 200 ml of medium, incubated in a rotary shaker Model Arthur Thomas, at 150 rpm at 30°C for 100 hours under fluorescent illumination with 13000 lux of intensity. Each experiment was conducted four times. They were carried out at different cell concentrations: 0.1, 0.5, 1.0 and 2.0 g/l. Cadmium chloride, was added to the growth medium in concentrations ranging from 0.0 to 12.0 mg/l.

Cadmium chloride solutions were prepared from a concentrated stock solution, autoclaved at 111°C for 15 minutes and diluted for use in the experiments.

*Quantifications*: Cell growth was measured turbidimetrically at 430nm, and related to algae dry weight through a calibration curve. Cadmium uptake was determined in the supernatant, after cell centrifugation at 200 rpm for 15 minutes followed by filtration in 0.47 μm Millipore membranes, by atomic absorption spectrophotometry using Varian Techtron Spectrophotometer, Model AA6. The spectrophotometer was previously calibrated with a series of cadmium solutions prepared from a concentrated Merck standard cadmium solution.

**RESULTS AND DISCUSSION**

It was found that cellular growth ceased after 45 hours in cadmium free medium, in all tested conditions, the highest cell concentration obtained being 4.0 g/l, for *Chlorella* cells (Figure 1). The same final biomass was reached more quickly when the initial inoculum was increased. A 2.0 mg/l cadmium concentration did not affect cell growth of *Chlorella* significantly. These results are not in agreement with the literature (1,12), where the inhibition of growth for *Chlorella* cells was reported at cadmium concentrations lower than 2.0 mg/l. Our report shows negligible effect on cell growth at this cadmium concentration, but with increasing metal concentration the growth rate is reduced; this way, only a 2.0 g/l initial inoculum achieves maximum cell growth in presence of 12.0 mg/l. These observed profiles follow certain information in the literature about cadmium toxicity on algal cells and its effect on cell growth. Otherwise, the results were surprising due to the high metal concentrations employed, partially affecting the growth of *Chlorella hominophora* cells. This is probably associated to the use of a culture medium providing optimum conditions for cell growth. The absorption curves for different cadmium concentrations are not similar. It appears that the amount of cadmium absorbed by a specific cell inoculum diminishes as the metal concentration increases. The absorption becomes less effective as a function of cell contamination due to the presence of the metal, although it is complete at the final stages of the process. It is very interesting to notice the instantaneous metal uptake by the cells, a fact already detected by us and other researchers (2, 6, 10).

The growth of *Scenedesmus quadricauda* in cadmium containing medium was inhibited in all metals concentrations tested (Figure 2). The cells were again more resistant to ions effect in the highest inoculum, but still showed the repressive effect of cadmium on the final biomass growth, at the test conditions.

Doyle et al. (3) studying bacteria and molds reported a negative influence in almost all microorganisms in the presence of cadmium at 5 μg/l. However, the growth of *Escherichia coli* and *Streptococcus faecalis* was stimulated with 5 and 10 μg/l of cadmium, respectively.

In parallel we could observe cadmium uptake from solution. This was complete up to 6.0 mg/l as a initial metal concentration, even with a decrease in cell growth. The time required for metal accumulation increases with the metallic content of the medium. This fact can be understood if the metallic ion does not promote total inhibition of the metabolic activity. In this case cell growth is obtained. On the other hand it will generate more binding sites for cadmium uptake, indicating that algal cell walls (4) and extracellular material (9), are efficient biosorptive agents. On the other
FIGURE 1 - "Chlorella hornosphaera" cell growth and cadmium uptake in presence of varying cadmium concentrations:
(A) 0.0 mg/l; (B) 2.0 mg/l; (C) 4.0 mg/l; (D) 6.0 mg/l; (E) 12.0 mg/l.

Initial cell concentrations:
- (0.1 g/l); Δ (0.5 g/l); ○ (1.0 g/l); □ (2.0 g/l).
hand, the *Scenedesmus quadricauda* strain has shown to be extremely sensitive to cadmium in this purpose.

Experiments carried out in this way provide a favourable indication of the potential applicability of the sorption process for cadmium containing industrial effluents clean-up by *Chlorella homosphaera* cells. These results were obtained for a preliminary evaluation of the feasibility of using green microalgae as metal sorbents. After being captured, the metals, concentrated in the cell components, can be efficiently eluted. This is an important feature both industrial clean-up and for the concentration of heavy metals.
RESUMO

Captação do cádmio e seu efeito no crescimento de Chlorella homosphaera e Scenedesmus quadricauda em condições laboratoriais.

O efeito tóxico e a captação de cádmio durante o crescimento de Chlorella homosphaera e Scenedesmus quadricauda foram estudados em diferentes concentrações iniciais de células (0,1 a 2,0 g/l). As células foram crescidas em meio de crescimento contendo cloreto de cádmio nas concentrações de 0,0 a 12,0 mg/l. O crescimento celular de Chlorella e Scenedesmus foi afetado pelo metal nas concentrações de 4,0 mg/l e 2,0 mg/l, respectivamente. Para as duas espécies estudadas, este efeito foi diretamente proporcional à concentração do metal e, inversamente proporcional à concentração celular.

Palavras-chaves: Absorção de cádmio, microalgas verdes.

REFERENCES


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