TOXICITY AND BINDING OF COPPER, ZINC, AND CADMIUM
BY THE BLUE-GREEN ALGA, Chroococcus paris

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Abstract. The toxic effects and accumulation of the heavy metals, Cd, Cu, and Zn by the sheath forming blue-green alga Chroococcus paris were investigated. All three of the metals were bound rapidly. Approximately 90% of the total amount of the added metal was bound within 1 min. Further significant binding occurred at a slower rate. The maximum metal binding capacity, as determined by filtration studies, was determined to be 53, 120, and 65 mg g⁻¹ dry algal weight for Cd, Cu, and Zn, respectively. Binding curves for the metals followed the Langmuir adsorption isotherm model. The amount of metal bound increased with increasing pH. Metal binding increased significantly when pH was increased from 4 to 7. Nearly all of the metal was found to be rapidly EDTA extractable. Metals were found to be increasingly toxic to growing cultures in the order, Zn, Cd, and Cu. All of the metals studied exhibited toxic effects at concentrations greater than 1.0 mg L⁻¹. The lowest concentrations used which showed detectable toxicity were 0.1 mg L⁻¹ for Cu and > 0.4 mg L⁻¹ for Cd and Zn.

1. Introduction

Increasing usage of heavy metals and their widespread dissemination in the aquatic environment has stimulated many studies on the effects of heavy metals on phytoplankton (Whitton and Say, 1975; Gadd and Griffiths, 1978; DeFillipis and Pallaghy, 1976a, b; Whitton, 1970; Hart and Scaife, 1977; Leland et al., 1979). Only a few studies have been concerned with the effects of heavy metal toxicity on blue-green algae (Henriksson and DaSilva, 1978; Stratton and Corke, 1979; Laube et al., 1980; Allen et al., 1980; Singh and Pandey, 1981). This group of algae are important bloom organisms in lentic waters where they become the dominant factor in minor element metabolism and dynamics (Boyd and Lawrence, 1966). Their importance in the trophic food chain is acknowledged, consequently basic information on metal toxicity and binding is of interest.

The specific objectives of this work were to (1) determine the short term binding capacity of Cd, Zn, and Cu by a sheath producing blue-green alga, (2) determine its short term metal concentrating ability, and (3) determine the toxicity of these metals to this organism.

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2. Materials and Methods

The organism used in this study was *Chroococcus paris*, a unicellular, sheath forming blue-green alga obtained as an axenic culture from Dr. Mary M. Allen, Wellesley College, Wellesley, Mass. U.S.A. Cultures were grown in BG-11 medium (Allen, 1968) with rotary shaking at 120 rpm and 100 foot candleless (1076 Lux) under cool white fluorescent lamps (Stanier et al., 1971) at 26 °C. Sterility was determined using nutrient agar spread plates incubated at 26 °C.

Biomass was determined by filtering cultures through Gelman glass fiber filters and drying filters at 70 °C for 1 hr. Optical density determinations at 450 nm using a Perkin-Elmer 200 spectrophotometer were also correlated with biomass and used in some studies (1 cm light path, Δ OD of 0.1 units = 55 mg L⁻¹ cell dry wt).

Metal binding (6 to 9 replicates per experiment) was determined on cells washed twice in deionized, distilled water after centrifugation at 3000X g and resuspended in a final volume of 50 mL. Metal solutions, added as Cd(NO₃)₂·4H₂O, Cu(NO₃)₂·3H₂O, and ZnSO₄·7H₂O were contacted with the washed cells, with stirring, for 15 min before filtration through a 0.45 μm Millipore membrane. The filtrate from triplicate analyses was adjusted to pH 2 and analyzed by atomic absorption spectrometry (Van Loon, 1980).

Growth inhibition studies were performed by adding log phase cells (15 to 16 mg L⁻¹ dry wt) to 50 mL of BG-11 medium containing the metals in triplicate 125 mL flasks. Flasks were incubated on a rotary shaker at 26 °C under continuous 100 ft candle (1076 Lux) illumination. Growth rates were determined spectrophotometrically at 450 nm.

![Fig. 1. Time course of Cu, Zn, and Cd binding by *C. paris*. Washed, living cells (15 mg dry weight) were suspended in 50 mL of a 2 ppm solution of the metal. Range about the mean ±2.5%.

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