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Cadmium Induced Potassium Efflux from *Scenedesmus quadricauda*

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Plants, algae and bacteria respond to heavy metal toxicity by inducing different enzymes, ion influx/efflux for ionic balance and synthesize small peptides such as poly(γ- glutamyl cysteinyl) glycines called phytochelatins (PCs) mainly consisting of glutamate, cysteine and glycine. These peptides bind metal ions and reduce toxicity (Reddy and Prasad 1990).

The uptake of metal ions comprises two phases (Norris and Kelley 1977). The first phase consists of a quick and nonspecific binding of the cations to negetively-charged membrane components located at the cell surface. The second phase consists of energy-dependent intracellular uptake of the metal ions. During uptake of Co^{2+} by yeast cells, an electroneutral 2:1 exchange with cell K^+ was found (Norris and Kelley 1977). Cd^{2+} uptake by yeast also caused loss of cell K^+, however, there was no electroneutral exchange of K^+ (Kessels *et al.* 1985). The molar ratio of K^+ released and Cd^{2+} accumulated by yeast in the initial stage of incubation is 22 and seems to be independent of the Cd concentration (Kessels *et al.* 1985). Disruption of the cell membrane of part of the cells, according to an all-or-none process, by Cd^{2+} may explain the disproportional loss of cell K^+ during Cd^{2+} uptake (Belde *et al.* 1988).

This paper examines the exchange of K^+ with Cd^{2+} uptake in *Scenedesmus quadricauda*, and whether it follows an electroneutral 2:1 exchange or an all-or-none process.

MATERIALS AND METHODS

S.quadricauda cells were cultured in Chu-10 medium (pH 8.5),

Send reprint requests to Dr.M.N.V.Prasad at the above address at 25 ± 2^0C, 16hr photoperiod (1400-1600 lux). Growth was measured as dry weight by filtering on to GF/C filters. Cells were washed with fresh medium and incubated with varying concentrations of cadmium chloride. Potassium efflux was estimated by flame photometer (Digisun Electronics, India) in the supernatants obtained at appropriate times by centrifugation. Dried cells were digested in a sulphuric acid and perchloric acid mixture and cadmium was estimated by atomic absorption spectrometry (Varian Techtron 1000, Australia).

RESULTS AND DISCUSSION

K^+ efflux was observed during Cd^{2+} uptake, and it increased with increasing Cd^{2+} uptake into the cells (Fig.1A and B). A dose-related K^+ efflux was observed when *Scenedesmus quadricauda* cells were treated with varying concentrations of cadmium chloride (Fig.2A). Total K^+ content of the cells was 745 mM/kg dry wt. It is known that cadmium causes injury to cell membrane of yeast, resulting in the release of low molecular weight materials and ions from the cells (Theuvenet *et al.* 1987; Kessels *et al.* 1985). Overnell (1975) reported the loss of cell K^+ in two species of marine algae, *Dunaliella tertilecta* and *Phaeodactylum tricornatum* after exposure to heavy metals. Ohsumi *et al.* (1988) showed the release of aspartate and glutamate in addition to K^+ in copper-exposed yeast cells.

K^+ efflux did not follow the electroneutral 2:1 exchange with Cd^{2+} in *S. quadricauda.* Cd^{2+} uptake to K^+ efflux ratio ranged between 22 and 32. At initial stages the ratio was 25 (10 min), 22 (20 min) and reached to 32 within first 30 minutes (Fig.2B). During longer incubation of up to 3 hours, *S.quadricauda* showed a clear drop in Cd^{2+} uptake to K^+ release ratio. It was observed in yeast that within the first few minutes of incubating cells with Cd^{2+}, the molar ratio of released K^+ and accumulated Cd^{2+} was 22 and was independent of the amount of cadmium chloride concentration in the medium (Kessels *et al.* 1985). However, no single relation existed between the amount of K^+ released and the cellular Cd^{2+} concentration during longer incubation of yeast cells with cadmium chloride (Kessels *et al.* 1985).

In *S.quadricauda*, it was observed that loss of potassium was more than electroneutral 2:1 exchange with cadmium. This may be considered to be symptomatic of membrane disruption. Deactivation of enzymes involved in the selective permeability of

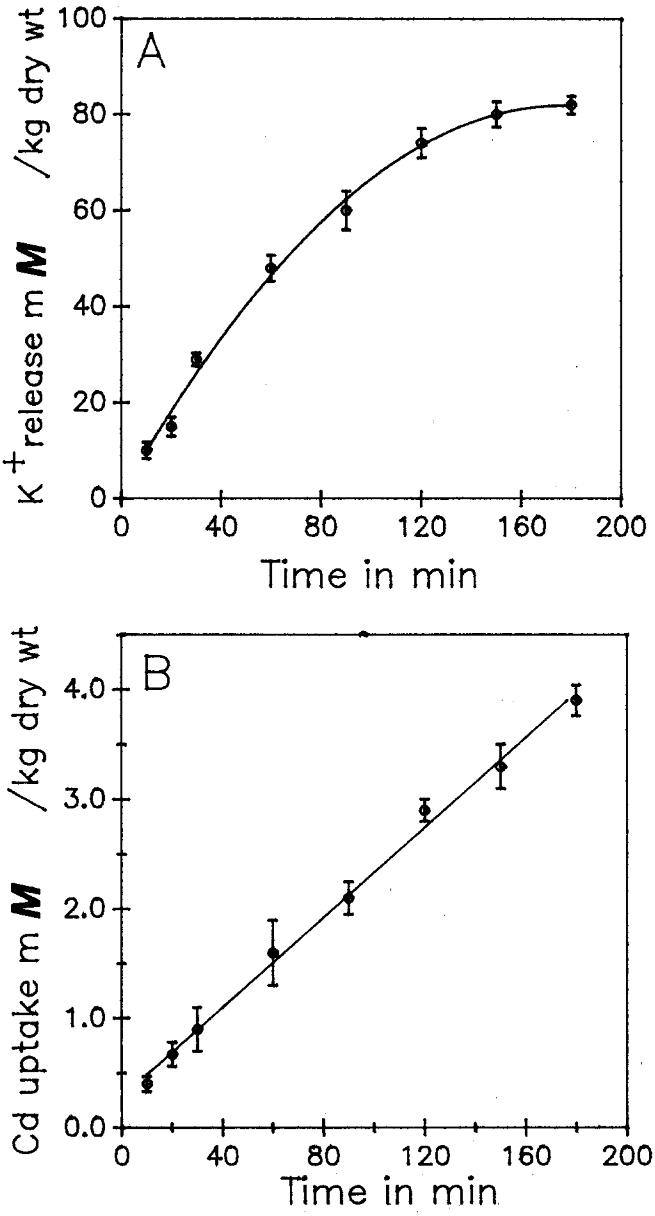

Figure.1A. Time course of K^+ release and Cd^{2+} uptake (1B) at 100μM external cadmium chloride in *S.quadricauda.*

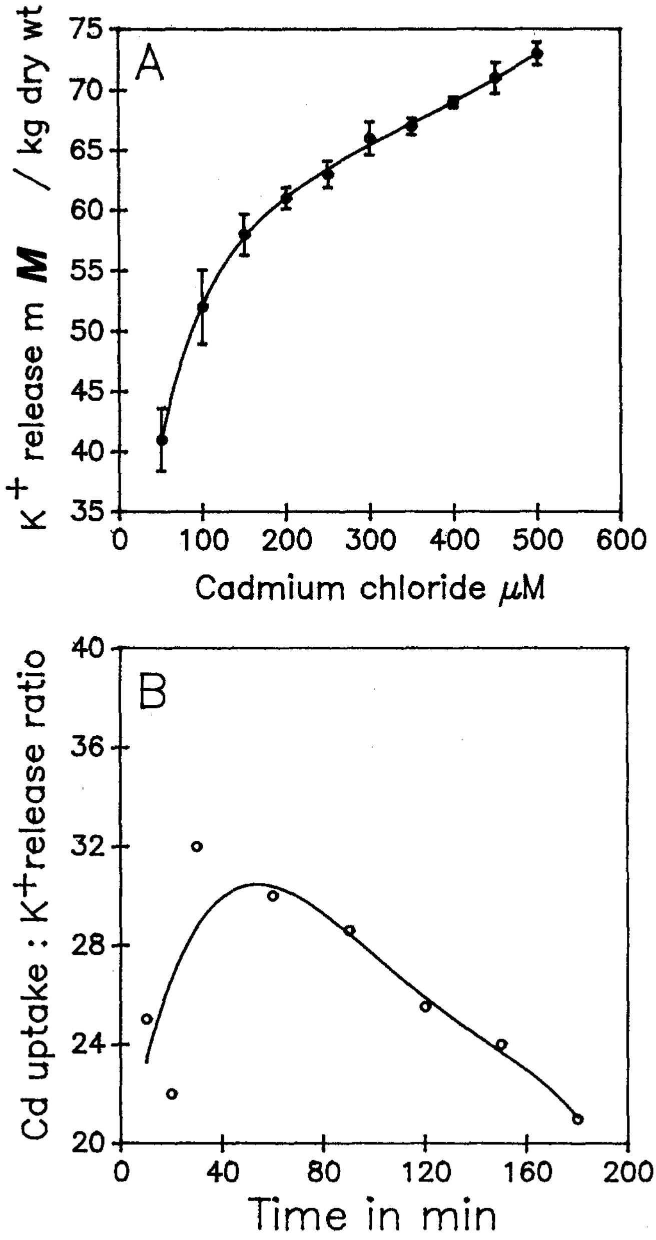

Figure.2A. *S.quadricauda* dose response curve of K^+ release and externally applied cadmium chloride. Total K^+ content of cells of 745 mM/Kg dry wt. 2B. Time course of Cd^{2+} uptake to K^+ release ratio in *S.quadricauda.*

membranes, such as ATPases, by copper was demonstrated by Veltrup (1982). In addition, it has been reported that the permeability of membranes depends on the degree of lipid peroxidation (Dhindsa *et al.* 1982). The lipid peroxidation process is believed to be initiated by free radicals such as the hydroxyl radical generated in the Haber-Weiss reaction, and peroxyl and alkoxyl radicals which are produced during the decomposition of organic hydroperoxides (De Vos *et al.* 1989). De Vos *et al.* (1989) reported copper-induced lipid peroxidation in *Silene cucubalus* and suggest that direct free radical formation leading to lipid peroxidation might play an important role in the mechanism of copper- induced damage to the permeability barrier in roots of higher plants *in vivo.* Cadmium-increased permeability of *S.quadricauda* may possibly be due to lipid peroxidation is to be investigated.

It is reported that heavy metals may provoke K^+ efflux from yeast in an all-or-none process (Belde *et al.* 1988). Each cell has according to this view a certain threshold concentration, which, if exceeded, results in the destruction of the permeability barrier for K^+. Consequently all the K^+ is released from the affected cells (Theuvenet *et al.* 1987). Kuypers and Roomans (1979) showed that the increased K^+ release from yeast exposed to increasing mercury levels was due to an increase in the number of cells showing an all-or-none response. K^+ loss from *S.quadricauda* is also of a similar manifestation.

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