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Cadmium, zinc and the uptake of calcium by two crabs, *Carcinus maenas* and *Eriocheir sinensis*

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Abstract

The uptake of dissolved cadmium and zinc by crustaceans can usually be explained by the passive process of facilitated diffusion involving a transport protein in the membranes of permeable surfaces. Cadmium ions will also enter via uptake routes for calcium, given the similar size of the two free ions. This study has investigated the interaction of cadmium (and comparatively zinc) and calcium uptake in two crabs that show different permeability responses to changes in salinity, with consequently different effects on the uptake of cadmium and zinc with salinity change. Ca uptake rates in *Carcinus maenas* decreased in reduced salinity (33–15) with the decreased Ca concentration of the medium and increased if the Ca concentration was increased at salinity 20. It is concluded that Ca uptake over the salinity range 33–15 is via apical Ca channels in gill ionocytes, passively down an electrochemical gradient. The Ca uptake rate of *Eriocheir sinensis* showed no significant decrease over the salinity range 33–10 (probably because of the small differences in an already low Ca uptake rate in this crab against a background of inter-individual variability), but decreased significantly at salinity 5. Added calcium increased the Ca uptake rate of *E. sinensis* at salinities 15 and 5, supporting the interpretation that Ca uptake in gills is typically passive via apical Ca channels. Cadmium (but not zinc) inhibited calcium uptake in both crabs at 15 salinity, indicating sharing of Ca channels by Cd, but not at salinity 5 (*E. sinensis* only) when Ca may be taken up into gill ionocytes by another (active?) physiological process.

Keywords: Uptake; Cadmium; Zinc; Calcium; Crabs; *Carcinus*; *Eriocheir*

1. Introduction

Marine invertebrates take up trace metals from solution across the permeable membranes of body surfaces in contact with the medium, probably by one or more of four possible transport routes: facilitated diffusion via a membrane carrier protein, a membrane channel consisting of a protein with a hydrophilic core, passive diffusion of non-polar metal species (e.g. $AgCl^0$), and by endocytosis (for example in the gills of bivalve molluscs) (Simkiss and Taylor, 1989, 1995; Tessier et al., 1994; Campbell, 1995; Rainbow, 1997). For many dissolved trace metals including Cd and Zn, the free metal ion is the model for the form of metal that is

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available for transport across the cell membrane via a carrier protein or through a membrane channel, and thus for the most bioavailable form of the dissolved trace metal (Tessier et al., 1994; Campbell, 1995). Physicochemical factors that affect the availabilities of these free metal ions in solution have correspondingly been shown to affect trace metal uptake rates by aquatic invertebrates (Nugegoda and Rainbow, 1988; O'Brien et al., 1990; Rainbow et al., 1993; Rainbow, 1997; Rainbow and Black, 2002).

The availabilities of free Cd and Zn ions in seawater are controlled by complexation by inorganic anions, particularly chloride (Bruland, 1983; Rainbow et al., 1993). Thus, dissolved Cd in seawater exists mostly as chloro-complexes with only 2.5% present as the free Cd^{2+} ion, while dissolved Zn is complexed by choride and hydroxide with about 47% present as the free Zn^{2+} ion (Zirino and Yamamoto, 1972; Mantoura et al., 1978; Bruland, 1983; Rainbow et al., 1993). A decrease in salinity reduces the amount of inorganic ions available for complexation of the dissolved metal and thereby increases the availability of the free metal ion. Thus, a decrease in salinity can increase the rate of uptake of dissolved Cd and Zn by an aquatic invertebrate simply via a physicochemical solution effect (Nugegoda and Rainbow, 1989a,b; Rainbow et al., 1993; Rainbow, 1997; Rainbow and Black, 2002).

Free cadmium ions are of almost the same ionic radius (0.92 Å) as calcium ions (0.94 Å) (Williams and Frausto da Silva, 1996), and it is inevitable that Cd ions will cross cell membranes via calcium channels (Simkiss and Taylor, 1995; Rainbow, 1997). Whether this is the main entry route for Cd, as opposed to facilitated diffusion via a membrane carrier protein, or even a significant entry route, will vary with the organism and its physiological state (Wright, 1977a,b, 1980; Rainbow, 1997). The rate of uptake of Cd by Ca channels has the potential to vary with physicochemical changes affecting calcium and/or cadmium ion concentrations in solution. Furthermore the activity of calcium pumps is under the physiological control of the invertebrate concerned, offering the potential for physiological intercession to affect a trace metal uptake rate (Rainbow, 1997).

Another, less metal-specific, physiological response that might affect trace metal uptake rates is the reduction in integumental permeability (measured as apparent water permeability (AWP)) shown by some, often euryhaline, invertebrates when salinity is reduced (Rasmussen and Andersen, 1996; Rainbow, 1997; Rainbow and Black, 2001), in correlation with reduction in Cd and Zn uptake rates from solution (Rainbow and Kwan, 1995; Rainbow and Black, 2002, in press). Thus, in the case of those invertebrates that have the ecophysiological ability to respond to a physicochemical change in solution such as a reduction in salinity, changes in the rate of uptake of cadmium and zinc will depend on the counterbalance between the opposing strengths of physiochemical enhancement of free ion availability and any physiological response, such as change in AWP, reducing trace metal uptake (Rainbow, 1997; Rainbow and Black, 2002).

Thus, the rates of uptake of Cd and Zn by the littoral amphipod crustacean *Orchestia gammarellus* follow the predicted increases in free metal ion concentrations on reduction in salinity down to 25, below which the amphipod appears to make a physiological response counteracting any further increase in Cd or Zn uptake rate with increased free metal ion availability (Rainbow et al., 1993; Rainbow and Kwan, 1995). In a more comprehensive study of cadmium and zinc uptake by three crabs, Rainbow and Black (2002, in press) have shown that the effect of reduced salinity on the rate of uptake of either metal is indeed controlled by the interaction of the physicochemical enhancement of uptake by increased availability of the free metal ion and the counteracting effect of physiological responses, probably including changes to AWP (Rainbow and Black, 2001) reducing trace metal uptake. Thus, the euryhaline crab *Carcinus maenas*, the common shore crab, and to a lesser extent the more stenohaline crab *Necora puber*, the velvet swimming crab, exhibited decreased Cd and Zn uptake from solution with reduction in salinity, the physiological response more than overcoming the physicochemical enhancement of free metal ion availability (Rainbow and Black, 2002, in press). In the case of the more euryhaline crab, the Chinese mitten crab *Eriocheir sinensis*, on the other hand, Cd and Zn uptake rates were increased with salinity reduction from 33 to 5, in line with physicochemical effects on the free metal ion availabilities, not (or little) affected by counteractive physiological responses (Rainbow and Black, 2002, in press). In fact the mitten crabs had a relatively unchanged AWP which was very low in comparison with the AWP of the other two crabs which reduced AWP with decrease in salinity (Rainbow and Black,