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Sex Specific Interactions Between Cd and Zn in Three Beetle Species (Insecta, Coleoptera)

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Cd and Zn are assumed to react in a similar way in biological systems (Nieboer and Richardson 1980) and it has been suggested that the mechanisms regulating the absorption of essential metals also regulate non-essential metals with chemical similarity (Turunen 1985). Soil dwelling arthropod species with high Zn concentrations often have high Cd concentrations as well (Janssen and Hogervorst 1993), indicating that these chemically related metals are regulated in similar ways. If this is the case, a correlation between Zn and Cd concentrations is likely to occur within insect species.

Female insects consume more food because of a larger reproductive investment. it has yet to be determined whether this larger consumption of food influences the ratio between Zn and Cd concentrations. Sex-specific accumulation of metals in carabid species have been reported (Rabitsch 1995, Lindqvist and Block 1997) but it is not known if this also reflects a difference in the ratio between different metals.

The aim of this study was to determine whether the relation between body concentrations of Zn and those of Cd are the same in male and female insects. For this purpose specimens of three beetle species with different life histories were collected and their concentrations of Zn and Cd measured. The correlation between Zn and Cd concentrations were then calculated for males and females. For one of the species specimen were analysed from two localities with different metal pollution loads.

MATERIALS AND METHODS

The species chosen for this study were *Pterostichus melanarius* Illiger (Carabidae) a predominately carnivorous species, especially in the adult stage (Lindroth 1986); *Geotrupes stercorosus* Scriba (Geotrupidae), feeds on animal faeces (Chinery 1973); and *Cetonia aurata* L. (Scarabaeidae), the adults of which visit various flowers and feed on pollen and nectar while the larvae live in decayed logs (Englund 1993).

The beetles were collected in April and May 1994 at Märsta in central Sweden. In addition, *P. melanarius* was also collected at Smedjebacken. This site is subjected

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to metal pollution caused by nearby factories, whereas no local source of metal pollution was present at Märsta.

The beetles were picked by hand and placed in a killing jar containing ethyl acetate. Fifty-one males and 30 females of *P. melanarius* were collected from Märsta, and 30 males and 18 females were obtained from Smedjebacken. Seventeen males and 14 females of *G. stercorosus* and 12 males and 13 females of *C. aurata* were caught. The beetles were stored in a freezer until the metal analyses were made. Prior to analyses they were lyophilized.

Due to the small amount of material in each individual, specimens of *P. melanarius* were pooled so that there were three individuals in each analysis. The other two species were analysed individually. The beetles were digested in test tubes of borosilicate glass. They were treated with 2 ml concentrated nitric acid of *pro analysi* quality at 110°C for 8 h. Thereafter they were heated in open tubes to near dryness. The residue was treated with 2-3 ml hydrogen peroxide (30%) of *pro analysi* quality at 110°C for 2 h to break down the remaining fat. Finally, the solutions were diluted with distilled water to five times the initial volume.

Metal analyses were carried out using atomic absorption spectrometry (AAS) with flame technique on a Shimadzu AA 670. Two reagent blanks to determine the baseline level were made for every 15 tests.

Correlations between concentrations of Cd and Zn were tested using the least squares method. Differences between metal concentrations were tested with Students T-test.

RESULTS AND DISCUSSION

Significant correlations ($p<0.05$) between Cd and Zn were found in females of all three species, while in males such a correlation was found only in *G. stercorosus*, fig 1. In this species a given increase in the level of Zn corresponded to a larger increase of Cd in males than in females. Also in the polluted area, Smedjebacken, a significant correlation ($p<0.05$) between Cd and Zn was found in the female but not in the male, fig 1 and 2. At this metal polluted site concentrations of Zn in males of *P. melanarius* were higher ($p<0.001$) than at Märsta, while no such difference was found for Cd. In females concentrations of both Zn ($p<0.05$) and Cd ($p<0.00l$) were higher in Smedjebacken.

Thus correlations between Zn and Cd concentrations occur for females in all the studied cases but for males only in one case. Metal concentrations in male insects can be affected by activities such as mating. The Zn concentration was found to decrease after mating in the male moth *Heliothis virescens* although this was not

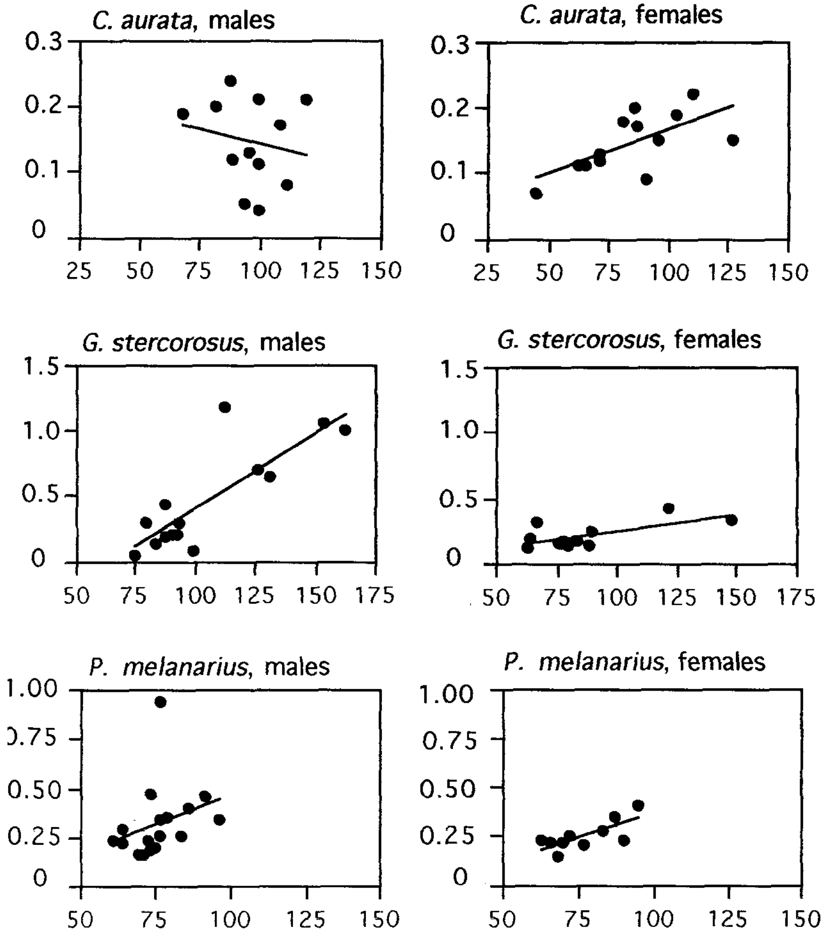

Figure 1. Concentrations of Cd (µg/g d.w.) are given on the y-axis. Zn (µg/g d.w.) concentrations are given on the x-axis. Equations for lines: *C. aurata* males: $y=-0.001x+0.23$, $r^2=0.03$ $p=0.63$, females: $y=0.001x+0.04$ $r^2=0.41$ $p<0.05$, *G. stercorosus* males: $y=0.012x-0.75$ $r_2=0.68$ $p<0.001$, females: $y=0.003x-0.01$ $r^2=0.48$ $p<0.01$, *P. melanarius* males: $y=0.006x-0.12$ $r^2=0.09$ $p=0.24$, females $y=0.005x-0.13$ $r^2=0.55$ $p<0.05$.

the case in the fly *Drosophila melanogaster* (Engebretson and Mason 1981). Mating in the studied species was assumed to take place during the collection period for *G. stercorosus* and *C. aurata,* but not for *P. melanarius.* It is therefore not likely that a decrease in the Zn level due to mating caused a lower correlation between Zn and Cd concentrations in males. This difference can be a result of different target organs. Cd concentrations differed between the sexes in the

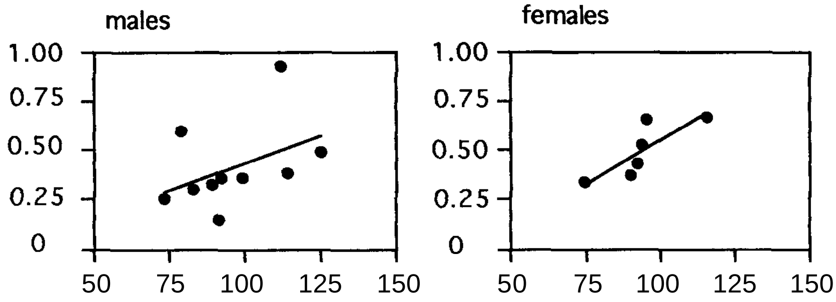

Figure 2. Concentrations of Cd (µg/g d.w.) in *P. melanarius* given on the y-axis. Zn (µg/g d.w.) concentrations are given on the x-axis. Equations for lines: males $y=0.006x-0.11, r^2=0.18$ $p=0.23$, females: $y=0.009x-0.31$, $r^2=0.67$ $p<0.05$

grasshopper *Aiolopus thalassinus* (Fabr.) owing to the fact that the target organs in males and females were not the same (Schmidt and Ibrahim 1994).

In the insect body Cd can replace other metals as co-factors in enzymes or bind to various other proteins. In these cases Cd can probably replace Zn and the ratio between Zn and Cd concentrations can be constant. On the other hand, Cd does not enter the body cavity to the same extent as Zn. In a study with *Pterostichus niger* Zn was found to be absorbed to a larger extent and distributed throughout the body, while Cd was mostly found in the gut epithelium or in connection with the integument (Lindqvist et al. 1995). Therefore it is not likely that the correlations found between Zn and Cd concentrations in the females are a result of uptake by the same mechanisms.

However, concentrations of Cd and Zn seem to influence each other in some way. In larvae of the silkworm *Bombyx mori*, Zn concentrations in the alimentary canal and Malpighian tubuli increased significantly with an increase in Cd concentration (Suzuki et al. 1984). In the beetle *Tenebrio molitor* (Vogel, 1988) and the cricket *Acheta domesticus* (Migula et al., 1989) Cd concentrations were found to be reduced by Zn. However, in a study with the collembola *Folsomia candida* Cd and Zn did not affect each other-s uptake (Cornelis et al., 1997).

In larvae of the fly *Drosophila melanogaster,* Cd initiated production of a cadmium-binding-protein, and most Cd was bound to this protein while Zn is not. Zn concentrations 100 times higher than that of Cd did not induce synthesis of a metal-binding protein, nor did it bind to the protein induced by Cd (Maroni and Watson 1985).

In the environment, Zn occurs in concentrations approximately 100 times those of Cd. Insects with a high Zn demand also assimilates a larger amount of Cd.

Although not taken up by the same mechanisms and not distributed in the same way in the insect body, a high uptake of Zn will cause a high uptake also of Cd.

The absorption and distribution of Cd and Zn in the insect body are dissimilar. Never the less, concentrations of Cd and Zn in females of all the studied species are linked to each other, although the mechanism is not known. In males the picture is unclear, and no overall pattern was found.

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