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Particle Size Selection in Cadmium Uptake by the Opossum Shrimp, *Mysis relicta*

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Fecal pellets of plankton play a key role in the recycling of trace contaminants within the aquatic environment (Benayoun 1974; Brown 1986; Martin et al. 1990). This is especially true for those plankton that undergo vertical migration in the water column. The opossum shrimp *Mysis relicta* is generally thought of as an omnivorous zooplankter occupying the pelagic zone of lakes, making itself available as a food source to a wide variety of fish. *M. relicta* undergoes diel migrations, meaning that a large proportion of its time is spent in the benthic regions of lakes feeding on sediments, particularly during the day (Lasenby and Langford 1973; Morgan and Beeton 1978; Morgan 1981). Van Duyn-Henderson and Lasenby (1986) found the concentrations of cadmium and zinc in the feces of *M. relicta* feeding on sediments to be 5-14 times higher than those of the surrounding food source. Several mechanisms have been suggested to explain the increase in concentration of metals from food source to fecal pellet. One possibility is that the lower assimilation efficiency of the metal compared to that of the organic fraction of the food may result in higher metal concentrations in the fecal pellet. It is also possible that, when feeding, organisms are selecting small particles associated with relatively high metal concentrations (Tessier et al. 1982; Mudroch and Duncan 1986), and inadvertently consuming more metal than is indicated by the average concentration detected in the bulk sediment (Brown 1986). This study examines the feeding behaviour of *M. relicta* to test the hypothesis that particle size selection can account for increased metal concentrations found in the fecal pellets of invertebrates.

MATERIALS AND METHODS

M. relicta were collected from Crystal Lake in southern Ontario (78° 19'W, 44° 45'N) in November of 1988. The organisms, feeding on zooplankton and sediments, were allowed to acclimatize for two weeks in 10 L aquaria containing aerated lake water kept at 5°C, in the dark.

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Spherical latex beads of various diameters (0.75-50 μm, Polysciences Inc.) were used for all particle size selection experiments. Three 10 μL subsamples were taken from each of ten beakers containing 100 mL of filtered lake water (0.45 μm) and a suspension of approximately 10^5 latex spheres of each size class (0.75, 2-19, 20-29, 30-39 and 40-50 μm). After the spheres were allowed to settle (5 hours) individual adult *M.relicta* (15.5-17.0 mm total length) were placed in the beakers for four hours, at 5°C, in the dark. At the conclusion of the feeding period the spheres within each beaker were resuspended (stirred with a glass rod) and three 10 μL subsamples were taken. All subsamples and *M. relicta* gut contents were mounted on glass slides with glycerine. The spheres within each slide were counted and categorized according to diameter (using an occular micrometer) at 400X magnification and again at 1000X magnification with an ultraviolet light source to view the fluorescent 0.75 μm particles. All bead counts were expressed as a percentage of the total number found on each slide.

The top 2 cm of thirteen sediment cores (K-B corer), collected from a 20 m depth in Crystal Lake, comprised the samples for cadmium (10 replicates) and grain size (3 replicates) analysis. For cadmium analysis the <63 μm size fraction was first separated using a wet sieve with acid washed plastic screening. Further separation (2-20 μm, 21-40 μm, 41-63 μm) was achieved using the settling column method (Holme and McIntyre 1971) in an acid washed Nalgene graduated cylinder. The separated sediment samples were then transferred into labelled, preweighed, acid washed teflon vials, dried at 125°C, and weighed. The digestion of each sediment sample for cadmium analysis required the addition of 2 ml of concentrated HNO_3, the application of heat (until the production of fumes had ceased), the addition of a 1 ml volume of concentrated HCL and further application of heat (until the production of white fumes had ceased). Finally, 2 ml of 0.1 N HNO_3 was added to each sample. All cadmium analysis was done by flameless atomic adsorption (Scintrex AAZ-2) equipped with Zeeman modulation. The accuracy of the analyses was checked by running three samples of standard reference material (NBS 1645). Standard hydrometer methods (Griffithe 1976) were used for the grain size analysis of the remaining three sediment samples.

RESULTS AND DISCUSSION

The feeding rates of *M. relicta* varied with the size range of particles offered. In the gut contents the .75 μm particles made up a significantly ($p < .05$) smaller percentage (2.85%) compared to that which was made available (19.32%) (Figure 1-a). The percentage of 2-19 μm particles in the gut of *M. relicta* (44.07%) was significantly ($p < .05$) higher than that within the beaker (31.12%). A comparison of the percentages of 20-29 μm and 30-39 μm spheres made available to those ingested by *M. relicta* revealed no significant differences. A significantly ($p < .05$) lower number of the largest particle size fraction (40-50 μm) was ingested (6.0%) compared to that made available within the beaker (14.21%).

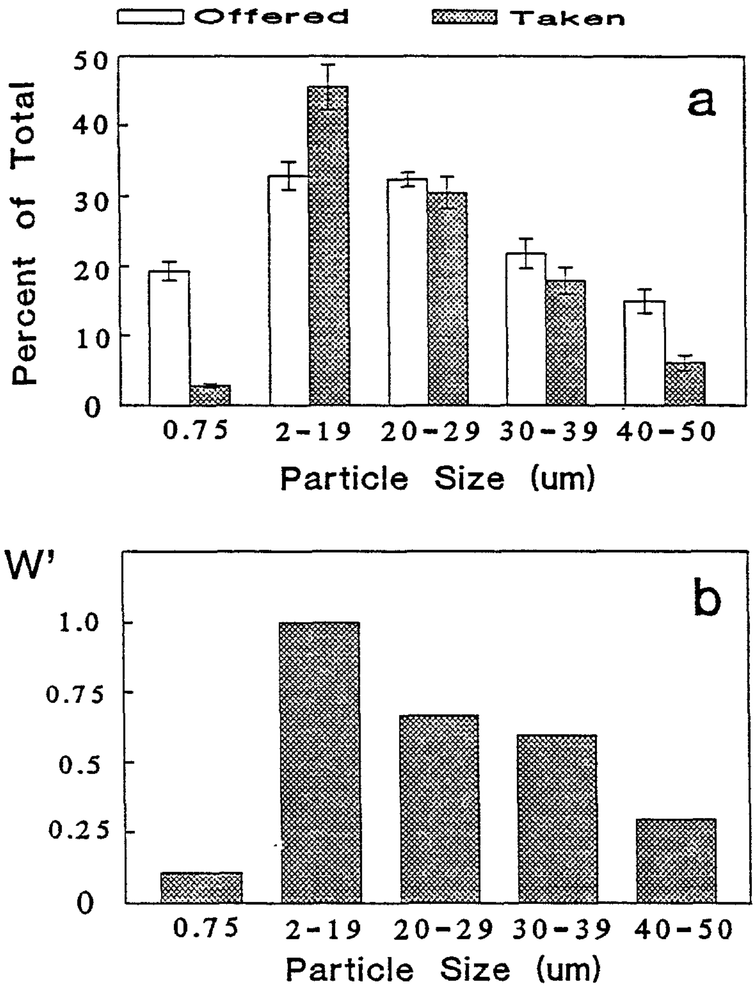

Figure 1. (a) The size distribution (µm) of particles in the gut contents of *M. relicta* (taken) and of particles available within the beaker (offered) (error bars represent standard deviations); **(b)** The selectivity indices for each particle size class ingested by *M. relicta* .

The polystyrene spheres, used to represent the sediment particles encountered by *M. relicta* feeding in the benthic region of lakes, provided a solution to the problem of fragmentation of particles upon ingestion (Bowers and Grossnickle 1978) and the subjectivity normally associated with making diameter measurements of natural sediment particles (due to the irregularities in shape). Similar spheres have been used successfully in the past to study seasonal resource partitioning patterns of marine calanoid copepods (Skiver 1980), in optimal forage modelling for deposit-feeding benthic invertebrates

(Taghon 1982), and in particle size selection experiments involving ciliates (Sanders 1988). Although Peters and Downing (1984) found this type of sphere to be ingested by copepods at lower rates than natural algal particles, they were ingested in the same size fraction proportions, and were therefore determined suitable for measuring size selection.

Figure 1-b expresses the results of the feeding experiment in the form of a selectivity index; W' (Vanderploeg 1981), calculated as:

$$W' = C_i / C_{pref}$$

where C_i is the clearance rate of particles of size *i* and C_{pref} is the highest clearance rate in the set of rates (i.e., the preferred size). The selectivity indices of Figure 1-b clearly show the preference toward particles of 2-19 μm. These results are consistent with the earlier observations of Lasenby and Langford (1973) who found that *M. relicta* feeding on sediment had a significantly higher percentage of particles in the range of 1 to 5 μm in the gut compared to that of the substrate, suggesting that the *M. relicta* may be more efficient at collecting these particular sizes. While indicating that *M. relicta* selects for small particles (2-19 μm), the results of the present study also suggest that there are upper (between 40 and 50 μm) and lower (between .75 and 2 μm) size limits of ingestible particles. The mid-size ranges of latex beads (20-39 μm) were ingested in the same proportions as were available in the beaker environment. Considering that Sprules et al. (1983) found that sediment particles of 6-9 μm diameters were present within 26 lakes studied in greater densities than the larger size fractions, the selection of small particles by *M. relicta* can be viewed as a potentially important phenomenon transferable to lake substrate conditions. Considering the present findings we hypothesized that by selectively feeding on the benthic sediments containing increased organics and higher cadmium levels *M. relicta* could be acting as a means for concentrating cadmium for subsequent uptake by other organisms, thereby participating in the cycling of this contaminant within the aquatic environment.

For Crystal Lake sediments a general decline in cadmium levels was found with increasing particle diameter (Table 1), with a significant difference ($p < .05$) determined between the cadmium concentrations of the 2-20 μm (2.09 μg Cd/g) and the 36-63 μm particles (1.27 μg Cd/g). The smallest grain size fraction analyzed for (2-20 μm) comprised 34.8% of the total sediment on average, while the 21-35 μm and 36-63 μm size fractions made up 18.7% and 46.5% of the total sediment, respectively (Table 1).

To determine the degree to which the selection of small sediment particles affects the cadmium uptake by *M. relicta*, and how it might compare with the concentration factors found by Van Duyn-Henderson and Lasenby (1986), estimates of cadmium ingested by both selective and non-selective *M. relicta* were made and compared (Table 1). The results of the grain size analysis performed on Crystal Lake sediment were used in combination with the cadmium-grain size data to determine the amount of cadmium ingested by a non-selective feeder. This value was then compared to the amount of cadmium ingested by a selective feeder. A non-selective feeder ingesting one mg of sediment would ingest 1.58 ng of cadmium while a selective feeder would ingest 1.70 ng. Therefore, selective *M. relicta* would take in 1.1 times the amount of cadmium that a non-selective feeder would. A maximum concentration factor would be experienced if *M. relicta* fed exclusively on the 2-20 μm size fraction where the amount of cadmium ingested would be 2.09 ng, 1.3 times that of non-selective *M. relicta*. These calculations suggest that particle size selection plays a minor role in the process of concentrating metals.

Table 1. Cadmium concentrations associated with the various sediment size fractions and calculations for the resulting amount of cadmium ingested by selective *M. relicta* compared to that of non-selective *M. relicta* feeding on sediment.

	Non-Selective *M. relicta*			Selective *M. relicta*	
Grain Size (μm)	Cadmium Conc. (ng/mg) **(A)**	Particle Size % Total **(B)**	Total Cd ingested (ng) **(AxB)**	Particle Size % Total **(C)**	Total Cd ingested (ng) **(AxC)**
2-20	2.09	34.8	0.73	46.0	0.96
21 - 35	1.42	18.7	0.26	39.0	0.55
36 - 63	1.27	46.5	0.59	15.0	0.19
TOTAL			1.58		1.70

When the rate of the assimilation of organic material relative to the rate of assimilation of cadmium is considered the concentration factor for cadmium in selective *M. relicta* increases. By applying the assimilation and ingestion rates for both sediment and cadmium to the particle size selection data of the present study the egestion rates for sediment and cadmium can be calculated as (Johannes and Satomi 1967):

$$(1)\ A = I - U$$
$$(2)\ U = I - A$$

where A = assimilation rate, I = ingestion rate and U = rate of release of unassimilated material.

Utilizing the ingestion and egestion rates from Table 2 the cadmium concentrations entering and leaving *M. relicta* can be calculated as such:

$$(3)\ \text{Ingested Cd Conc (ng/ug)} = I_c / I_s$$

$$(4)\ \text{Egested Cd Conc (ng/ug)} = U_c / U_s$$

where I_c = the ingestion rate for cadmium (.043 ng/hr), I_s = the ingestion rate for sediment (15.5 μg/hr), U_c = egestion rate for cadmium (.047 ng/hr) and U_s = the egestion rate for sediment (3.1 μg/hr).

Table 2. Calculated egestion rates of cadmium and sediment for *M. relicta* using equation (2) above.

	I	Corrected I	A	Calculated U
Sediment	15.5 ug/hr[1]	----	12.4 ug/hr[2]	3.1 ug/hr
Cadmium	.043 ng/hr[1]	.047 ng/hr[3]	.00039 ng/hr[2]	.047 ng/hr

[1] Van Duyn-Henderson 1985
[2] Lasenby, *unpublished*
[3] .043 ng/hr x 1.1 (concentration factor for selecting *M. relicta*)

The ingested cadmium concentration of .00277 ug/ng (equation 3) and an egested cadmium concentration of .0152 ng/ug (equation 4) result in a concentration factor of 5.5 which is in the middle of the range found by VanDuyn Henderson and Lasenby (1986) for cadmium (2-9.7).
These results illustrate the importance of assimilation efficiencies in resulting metal concentrations. Although the results of this study indicate that particle size selection is not an important factor in determining the high cadmium concentrations found for *M. relicta* feeding on sediments, this feeding pattern may have important implications for the ingestion, and subsequent recycling, of other contaminants. For example, Brown (1986) found that copper, zinc, and nickel concentrations increased 2-4 times in sediment particles decreasing from 125 to <63 μm. For metals, such as these, with a more pronounced pattern of increasing concentration with decreasing particle size, the particle size selection hypothesis may play a more prominent role in the cycling of trace contaminants in aquatic ecosystems.

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