

Changes in the Amount of Cadmium in Food Ingested by the Creek Chub, *Semotilus atromaculatus*

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The importance of food-borne cadmium in determining the cadmium body burdens of fish depends on a variety of factors such as the fish species and the food type. Williams and Geisy (1978) reported no uptake of Cd by the mosquito fish, *Gambusia affinis*, from Cd-spiked flake food. Similarly, Singh and Ferns (1978) found no gastrointestinal absorption of Cd by rainbow trout, *Salmo gairdneri*, fed a prepared food containing sewage sludge with its associated heavy metals. Alternatively, guppies, *Poecilia reticulata*, fed Cd-spiked *Daphnia* did accumulate cadmium (Hatakeyama and Yasuno 1982) as did rainbow trout, *S. gairdneri*, fed a prepared diet (Kumada *et al.* 1980). Of these researchers, those reporting no gastrointestinal uptake of Cd used foods containing Cd at concentrations several orders of magnitude lower than those who do report gastrointestinal absorption of the metal (Table 1).

The gastrointestinal tract of fish has also been implicated in the excretion of cadmium. Increased metal content of fecal matter of fish experimentally exposed to cadmium in solution has been reported (Harrison and Klaverkamp 1989), as well as apparent cadmium enrichment of prey items as they travel through the fish's gastrointestinal tract (Dallinger and Kautzky 1985; Amiard *et al.* 1980; Amiard – Triquet *et al.* 1980). The presence of metal-binding corpuscles in the gastrointestinal lumen of some marine fish species (Noel-Lambot 1981) also suggests that the gastrointestinal tract may play a role in the elimination of cadmium by fish.

Our study examines the change in cadmium concentration and mass of cadmium in food before and after entering the gastrointestinal tract of a fish. Larvae of the blackfly family, Simuliidae, were used as the food source and the creek chub *Semotilus atromaculatus* as the test fish.

MATERIALS AND METHODS

Blackfly larvae used as food were collected from streams in the Peterborough area of Ontario, Canada in early April when the larvae were most plentiful. After

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rinsing them free of any debris, the larvae were spiked with Cd by adding $\text{Cd}(\text{NO}_3)_2$ to their holding water at a concentration of 2 mg/L which was also vigorously aerated to create the current to which the larvae are accustomed in the streams. Exposure time to the Cd-spiked water was altered from 5 to 48 hr to achieve different Cd concentrations in the larvae. Spiked larvae were rinsed with tap water for ½ hr, frozen, and 5 to 6 subsamples of the frozen larvae were analyzed for cadmium to determine the concentration of Cd in the food.

Creek chub were placed in individual compartments of 100 l aquaria which had been divided equally into 5 sections and maintained at 22°C. They were fed unspiked food, regularly, until they were observed to consume all the food offered to them in a short period of time, and were then starved for 2 d in preparation for the feeding experiment. For each trial, 5 to 9 starved fish were each fed approximately one gram wet weight (155 to 419 mg dry weight) of blackfly larvae of a known Cd concentration and allowed to digest the food for 8 hr, a time period less than their gut passage time. Fish were then sacrificed and their entire gastrointestinal tracts removed. The ingested blackfly larvae were removed from the tracts using clean acid-washed plastic forceps to gently squeeze the material out. All of the material was removed regardless of location in the tract or state of digestion. Gut contents were placed in acid-washed Teflon vials, dried and weighed.

The dried samples were dissolved with 2 ml of concentrated nitric acid (Baker Analyzed Reagent) and heated on a hot plate for 2 hr at 100°C. The samples were then allowed to cool and 1 ml hydrogen peroxide (30% by vol., Fluka Chemie) added. The ensuing bubbling reaction was allowed to subside and the samples were evaporated to approximately 0.1 ml. The samples were then diluted to 4 ml with distilled, deionized water. Cadmium analysis was done on a Scintrex AAZ 2, Zeeman-modulated, flameless atomic absorption spectrophotometer equipped with a tungsten filament. To check accuracy and precision, samples of NIST standard reference Bovine Liver Tissue (NIST #1577a) were run and yielded concentrations within the certified value of $0.44 \pm .06 \mu\text{g Cd/g dry weight}$. Percent recovery of cadmium was greater than 97%.

Relative assimilation efficiencies of the fish at the different feeding levels and food cadmium concentrations were estimated by determining the difference between dry weight of food ingested and dry weight of food in the gut as a percentage of the dry weight ingested. These are not actual assimilation efficiencies as the food had not passed through the entire gut.

RESULTS AND DISCUSSION

Blackfly larvae had a much higher cadmium concentration after ingestion by the creek chub than before ingestion (Table 2). This difference became less pronounced with higher cadmium concentrations and was reversed at the highest concentration of 18,320 ng/g. Similar increases in metal concentration of ingested

Table 1. Cadmium concentrations of food reported in gastrointestinal-cadmium uptake studies in various fish species.

Species	Food type	Food Cd conc. ng/g	Uptake Yes/No	Reference
Guppy	<i>Daphnia</i>	69,500	yes	Hatakeyama and Yasuno 1992
"	"	125,000	yes	"
"	"	170,610	yes	"
"	"	560	no	"
Mosquito fish	flake food	115	no	Williams and Geisy 1978
"	"	1130	no	"
Rainbow trout	prepared food, 30% sewage sludge	1100	no	Singh and Ferns 1978
"	prepared food	1000	no	Kumada et al. 1980
"	"	10,000	yes	"
"	"	100,000	yes	"

food has been observed by others. In an attempt to determine the possibility of metal biomagnification in an estuarine food chain, Amiard *et al.* (1980) and Amiard - Triquet *et al.* (1980) found that cadmium concentrations of predator stomach contents were higher than the cadmium levels of prey before consumption. Further, the intestinal contents of the predators had higher cadmium concentrations than either the stomach contents or the prey items such that; prey concentration < stomach contents < intestinal contents. The high cadmium concentration in the intestinal contents was attributed to unassimilated metal enrichment in the feces. Preferential absorption of nutrients over metal ions was suggested as the underlying mechanism.

When total cadmium present is taken into account, the amount of cadmium present in the food in the gut of the creek chub was found to be from 1.3 to 2.4 times that originally ingested. This increase was only observed when cadmium concentrations of the larvae were low (431 and 580 ng/g) and ingested food levels were high. The only possible source of this "extra" cadmium was the fish itself. The results suggest that at low, natural cadmium levels, gastrointestinal elimination into food may represent a means of excretion of cadmium for the creek chub. The presence of corpuscles with a high cadmium binding capacity in

Table 2. Cadmium concentration and actual cadmium content of blackflies before and after ingestion by the Creek chub, *Semotilus atromaculatus*. (\pm s.e., A.E. = assimilation efficiency)

Dry wt ingested (mg)	[Cd] in food (ng/g)	Cd ingested (ng)	Dry wt in gut (mg)	[Cd] in gut (ng/g)	Cd in gut (ng)	n	A.E. %
414	431 \pm 47	181	146	2985 \pm 393	416 \pm 49	9	67
419	580 \pm 53	243	140	2186 \pm 291	305 \pm 53	7	64
155	1426 \pm 47	221	35	2046 \pm 698	54 \pm 9.9	5	77
166	18320 \pm 1447	3033	18	16060 \pm 1520	297 \pm 33	5	89

the intestinal lumen of various marine fish has been suggested as a possible mechanism of cadmium tolerance by Noel-Lambot (1981). The corpuscles, regularly evacuated through the anus, were thought to scavenge cadmium from imbibed seawater. The author hesitated to suggest that the corpuscles may be able to scavenge metals across the gastrointestinal wall of the fish. Although no attempt was made to find similar corpuscles in the creek chub, they may represent a possible mechanism of excretion in this species.

When fish were fed larvae with cadmium concentrations of 1,426 and 18,320 ng/g considerably less cadmium was found in the intestinal tract than was originally ingested.

The results of our study suggest that there is a threshold cadmium concentration in food below which no decrease of cadmium occurs and above which a decrease of cadmium does occur in the food in the gut. This threshold, at least for creek chub, appears to be between 580 and 1426 ng/g. Generally, researchers using food concentrations below this threshold claim no uptake from food sources, while those using concentrations higher than the threshold suggest that gastrointestinal absorption of cadmium is important (Table 1). The bioavailability of cadmium, however, may also determine whether or not gastrointestinal absorption takes place. At the higher concentrations, most of the cadmium associated with the larvae may have been adsorbed to their surfaces. This surface adsorbed cadmium may have been easily removed by the acid pH of the creek chub gut and assimilated by the fish. It is also possible that the cadmium lost from the food in our study may be in liquid form in the gut.

The amount of cadmium lost from the food may also be related to the amount of food ingested. The ingestion of greater amounts of lower-cadmium-concentration

food resulted in lower assimilation efficiencies and increases in the amount of cadmium in the same material in the gut. The ingestion of smaller amounts of food with higher cadmium concentrations resulted in higher assimilation efficiencies and decreases in the amount of cadmium in the gut. The effect of the amount of food ingested on the assimilation of cadmium and, possibly, other trace metals warrants further study.

From an ecological perspective, the importance of food as a cadmium source to fish will undoubtedly vary according to the species in question and other variables such as feeding rates. There does appear to be a threshold cadmium concentration of the food of between 580 and 1,460 ng/g dry weight below which no loss of cadmium from the food occurs and above which cadmium is lost from the food and possibly absorbed through the intestinal wall of the creek chub for the specific food type as labelled in this experiment. At low natural cadmium concentrations found in the blackfly larvae, their consumption may represent a means of cadmium excretion for the creek chub and, possibly, other fish.

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