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Metals in mandibles of stored product insects: do zinc and manganese enhance the ability of larvae to infest seeds?

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Abstract

Although high concentrations of zinc and manganese were found in mandibles of insect larvae that bore into seeds, these metals were not detected in mandibles of insect larvae that attack previously damaged seeds. Metals were present in the larval mandibles of a lepidopteran, the Angoumois grain moth (*Sitotroga cerealella*), and eight coleopterans, the lesser grain borer (*Rhyzopertha dominica*), cigarette beetle (*Lasioderma serricorne*), drugstore beetle (*Stegobium paniceum*), spider beetle (*Gibbium aequinoctiale*), warehouse beetle (*Trogoderma variabile*), cadelle (*Tenebroides mauritanicus*), larger black flour beetle (*Cynaeus angustus*), and cowpea weevil (*Callosobruchus maculatus*). Larvae of these species can chew into seeds. Larvae of six other coleopterans, the varied carpet beetle (*Anthrenus verbasci*), sawtoothed grain beetle (*Oryzaephilus surinamensis*), rusty grain beetle (*Cryptolestes ferrugineus*), red flour beetle (*Tribolium castaneum*), longheaded flour beetle (*Latheticus oryzae*), and granary weevil (*Sitophilus granarius*) have little if any ability to chew into seeds, and did not have metal in their mandibles. Larvae of the granary weevil hatch and feed within seeds that were penetrated previously during egg deposition by adults. However, newly hatched larvae of the cowpea weevil and the Angoumois grain moth have to bore through the seed coat before they begin feeding, and they have mandibles with high concentrations of zinc. These data support the hypothesis that deposition of zinc and/or manganese in larval mandibles enhances the larva's ability to penetrate seeds. Published by Elsevier Science Ltd.

Keywords: Mandible; Cuticle; Zinc; Manganese; Metal; Stored product; Grain; Seed; Insect; Beetle; Moth

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1. Introduction

High concentrations of metals are correlated with extreme hardness of the insect cuticle (Hillerton et al., 1982; Hillerton and Vincent, 1982; Edwards et al., 1993). Metals such as zinc and manganese are abundant in the cutting edges of the mandibles of adult beetles that attack seeds (Hillerton et al., 1984). High concentrations of metals occur in localized areas of the mouthparts of many arthropods, such as the tips of the fangs of a spider, which have as much as 15% zinc (Schofield and Lefevre, 1989).

When structures do not need to be hard and durable, they may lack metals. For example, the ovipositors of some species of wasps that place their eggs within soft substrates lack metal, while the ovipositors of species that must bore through harder substrates have metal (Quicke et al., 1998). The occurrence of metal in the mandibles of stored product insects could be more associated with feeding habits in the larval stages than in the adult stage, since larvae acquire new mandibles each time they molt, and thus they may have less need for durable mandibles. Surprisingly, with one exception, the occurrence of metals in larval mandibles of stored product insects has not been investigated. No metals were detected in the larval mandibles of the yellow mealworm, *Tenebrio molitor* L., although manganese was present in the adult mandibles (Hillerton and Vincent, 1982). The adults of this tenebrionid feed for several months, and both the adults and larvae usually feed on relatively moist seeds that have been broken or damaged (Arbogast, 1991; Sinha and Watters, 1985).

The presence of zinc and manganese in an insect's mandibles may be related to factors such as its feeding habits, phylogenetic position, and ability to acquire enough of these metals during feeding. To determine whether metal-enrichment of mandibles is an important characteristic of species that chew into seeds, we recorded the presence of metals in larval and adult mandibles of several species that differ greatly in their ability to bore through the outer seed coat. We also investigated whether dietary supplements of manganese or zinc may influence the deposition of these elements in the mandibles of one of the species.

2. Materials and methods

2.1. Insects

Insects were obtained from stock cultures maintained at the ARS Grain Marketing and Production Research Center at 25°C and ~60% r.h. The cigarette beetle, *Lasioderma serricorne* (F.), drugstore beetle, *Stegobium paniceum* (L.), spider beetle, *Gibbium aequinoctiale* Boieldieu, red flour beetle, *Tribolium castaneum* (Herbst), and longheaded flour beetle, *Latheticus oryzae* Waterhouse, were reared on a mixture of 95% wheat flour and 5% brewer's yeast. The cadelle, *Tenebroides mauritanicus* (L.), was raised on 95% whole-wheat flour and 5% brewer's yeast. The sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.), and rusty grain beetle, *Cryptolestes ferrugineus* (Stephens), were reared on 90% rolled oats, 5% brewer's yeast, and 5% wheat germ. The warehouse beetle, *Trogoderma variabile* Ballion, was cultured on 50% rolled oats and 50% powdered dog food (a lamb and rice formulation from Purina). The varied carpet beetle, *Anthrenus verbasci* (L.), was reared on dog hair and a mixture of wheat germ, honey, glycerol, and mold inhibitors (McGaughey, 1985). The larger black flour beetle, *Cynaeus angustus* (LeConte), was raised on cracked corn. The cowpea weevil, *Callosobruchus maculatus* (F.), was reared on cowpea. The Angoumois grain moth, *Sitotroga cerealella* (Olivier), and granary weevil, *Sitophilus granarius* (L.), were cultured on wheat kernels. The lesser grain borer, *Rhyzopertha dominica* (F.), was reared on wheat germ or corn meal (see below).

2.2. Effect of dietary supplements

Raw wheat germ flakes or corn kernels were ground through a screen (8 mesh/cm) in a Wiley Mill. The powdered wheat germ was then ground in a mortar with a pestle, either alone or with the addition of 300 ppm manganese ions (5.5 μmol/g) or zinc ions (4.6 μmol/g) as ACS grade sulfates ($MnSO_4 \cdot H_2O$ or $ZnSO_4 \cdot 7H_2O$) which were obtained from Fisher Scientific (Fair Lawn, NJ). *Rhyzopertha dominica* was placed on these diets at 28°C and ~75% r.h. Each individual was reared on 8 mg of diet in a 0.2 ml capillary pipette tip with the capillary end sealed. The larval exuvium that had remained attached to the posterior end of the pupal exuvium was saved and kept in association with the adult. Larval and adult mandibles were examined from each of the individual insects, and the Fisher exact test was used to test for significant differences between numbers of individuals that contained or lacked manganese in both sets of mandibles.

2.3. Metal analysis

Insects that were fully pigmented and sclerotized were killed by freezing and then lyophilized and mounted on adhesive carbon pads attached to aluminum stubs. At The Natural History Museum (London), the secondary and back-scattered images for carbon-coated specimens were obtained on a Hitachi S2500 scanning electron microscope (SEM) operating between 10 and 25 kV with a working distance of 35 mm. Metals were detected using a LINK AN10000 energy-dispersive analyzing system as described by Quicke et al. (1998).

The distribution of metals in the cuticle was mapped. Then the enrichment in specific locations was quantified using the peak to background method recommended by Roomans (1988). The abundance of zinc and manganese was recorded at a location that was likely to be enriched, such as a tooth or close to the cutting edge of a mandible. Background abundance was simultaneously recorded at a location on another area of sclerotized cuticle (on the head or, in large insects, near the base of the mandible). Enrichment was recorded as the ratio of the amounts detected at these two locations. This procedure compensates for instrumental factors that affect the counting rate and also for the absorption of X-rays by the specimen.

Larvae of *T. castaneum* and *C. angustus* were also examined at Kansas State University. After the labium and maxillae were removed, the insect was lightly coated with gold and palladium. The ventral surface of the mandibles was examined with a Hitachi model S-3500N SEM that was equipped with an Oxford model 7021 detector for energy dispersive X-ray analysis. The peaks from the gold and palladium coating were deleted from the analysis by using the Oxford Inca Energy Microanalysis software, and the concentration of manganese was estimated by comparing its abundance to the amount of other detectable elements that always included carbon and oxygen. These estimates are only semi-quantitative, because accurate quantitation would require the use of standards in an organic matrix similar to that of the specimen (Roomans, 1988). When attempting to detect the presence of a peak for manganese in the X-ray emission spectrum, similar numbers of counts were recorded from each sample point.

3. Results

Mapping of the abundance of metals in the insect specimens demonstrated that enrichment occurred in localized areas, typically on the cutting edges or teeth of the mandibles. After the preliminary mapping, enrichment was recorded as ratios of abundance in localized areas to the background abundance in another area of sclerotized cuticle. During interpretation of data on enrichment, any statements about the presence of metals are actually statements about the presence of enrichment, which for our study was defined as being a ratio of $\geqslant 3$ for zinc or a ratio of $\geqslant 1.5$ for manganese.

Enrichment with zinc was detected in mandibles of seven species of stored product insects (Table 1). It was detected in each stage of development that was examined in these species. The first instar larvae of two species (a moth, *S. cerealella*, and a chrysomeloid beetle, *C. maculatus*) were examined, because they bore into seeds. High levels of enrichment were found in the mandibles of these first instar larvae, and in the mandibles of the last instar larvae of *C. maculatus*. Enrichment was also detected in the mandibles of the one adult of *C. maculatus* that was examined. Therefore, in *C. maculatus*, enrichment occurred in mandibles of at least three stages, the first instar larva, the last instar larva, and the adult.

With the exception of the larvae of *C. maculatus and S. cerealella*, all of the larvae that we examined were in the last instar. Enrichment with zinc occurred in the larval mandibles of a cleroid beetle (*T. mauritanicus*) and four of six species of bostrichoid beetles (*R. dominica*, *L. serricorne*, *S. paniceum*, and *G. aequinoctiale*). The bostrichoid beetles from the Dermestidae (*A. verbasci* and *T. variabile*) did not have zinc in their mandibles. No enrichment with either zinc or manganese occurred in mandibles of the larval or adult stage of *A. verbasci*. Manganese was present, however, in the larval and adult mandibles of *T. variabile*. Manganese was not present in the non-functional pupal mandibles of *T. variabile*.

Zinc was not present in the mandibles of cucujoids, tenebrionoids, and curculionoids that were examined (Table 1). Manganese was detected in adult mandibles of these superfamilies and was typically found in larval mandibles of a tenebrionoid (*C. angustus*). Four out of five larvae of *C. angustus* had mandibles enriched with manganese. In contrast, only one out of 13 larvae of *T. castaneum* had mandibles enriched with manganese. None of the larvae of *L. oryzae*, *O. surinamensis*, *C. ferrugineus*, and *S. granarius* had mandibles enriched with manganese.

We examined additional specimens of *T. castaneum* and *C. angustus* using different equipment for the analysis (see Materials and methods section) to determine whether there was a consistent difference in the occurrence of manganese in these species. Peaks for manganese were not detected in the X-ray emission spectra for any of the sample points on the mandibles of three larvae of *T. castaneum*, although peaks for manganese were detected for similar sample points on all three larvae of *C. angustus*. The methodology provided semi-quantitative estimates of the concentrations. Manganese accounted for as many as 0.08%, 0.09%, and 0.83% of the atoms in the mandibular teeth of the specimens of *C. angustus* but was not detected near the base of the

Table 1
The presence of zinc and manganese in mandibles of stored product insects[a]

Taxonomy	Stage (n)	Metal content (range)	
		Zn	Mn
Lepidoptera			
Gelechiidae			
Sitotroga cerealella	Neonate larva (2)	4	0
Coleoptera, Chrysomeloidea			
Chrysomelidae, Bruchinae			
Callosobruchus maculatus	Neonate larva (5)	4 (3–5)	0
	Larva (4)	3 (1–5)	0 (0–1)
	Adult (1)	1	0
Coleoptera, Curculionoidea			
Curculionidae			
Sitophilus granarius	Larva (3)	0	0
	Adult (2)	0	2
Coleoptera, Cleroidea			
Trogossitidae			
Tenebroides mauritanicus	Larva (2)	3	0
Coleoptera, Cucujoidea			
Silvanidae			
Oryzaephilus surinamensis	Larva (4)	0	0
	Adult (4)	0	1 (0–2)
Laemophloeidae			
Cryptolestes ferrugineus	Larva (5)	0	0
	Adult (1)	0	4
Coleoptera, Tenebrionoidea			
Tenebrionidae, Diaperinae			
Cynaeus angustus	Larva (5)	0	1 (0–2)
	Adult (1)	0	1
Tenebrionidae, Tenebrioninae			
Tribolium castaneum	Larva (13)	0	0 (0–1)
	Adult (3)	0	1
Tenebrionidae, Tenebrioninae			
Latheticus oryzae	Larva (2)	0	0
	Adult (3)	0	1 (0–1)
Coleoptera, Bostrichoidea			
Dermestidae, Megatominae			
Anthrenus verbasci	Larva (2)	0	0
	Adult (2)	0	0
Dermestidae, Megatominae			
Trogoderma variabile	Larva (4)	0	3 (2–4)
	Pupa (2)	0	0
	Adult (2)	0	5

Table 1 (*continued*)

Taxonomy	Stage (*n*)	Metal content (range)	
		Zn	Mn
Anobiidae, Xyletininae			
Lasioderma serricorne	Larva (2)	1	0
	Adult (2)	2 (1–3)	1
Anobiidae, Anobiinae			
Stegobium paniceum	Larva (4)	2 (1–5)	1 (0–1)
	Adult (2)	1	1 (0–1)
Anobiidae, Ptininae			
Gibbium aequinoctiale	Larva (4)	2 (1–2)	1 (0–1)
	Adult (2)	1	2 (1–2)
Bostrichidae			
Rhyzopertha dominica			
Larval exuviae from 4 dietary regimes			
Corn meal	(6)	1 (0–2)	1 (0–1)
Wheat germ	(6)	2 (1–2)	1 (0–1)
Wheat germ + Zn	(3)	1 (1–2)	1
Wheat germ + Mn	(3)	2 (2–3)	1 (1–2)
Adults from 4 dietary regimes			
Corn meal	(6)	1 (1–2)	0 (0–1)
Wheat germ	(6)	1 (1–2)	0 (0–1)
Wheat germ + Zn	(3)	1 (1–2)	1 (0–1)
Wheat germ + Mn	(3)	1 (1–2)	1 (1–2)

[a] The enrichment with metal was scored (0) when no enrichment was detected relative to an unenriched area of sclerotized cuticle. The enrichment scale for zinc was (1) 3–6 $\times$ (i.e. the concentration was 3–6 $\times$ that in unenriched cuticle), (2) 7–10 $\times$, (3) 11–14 $\times$, (4) 15–18 $\times$, (5) $\geqslant 19\times$, and the scale for manganese was (1) 1.5–2 $\times$, (2) 2.5–3 $\times$, (3) 3.5–4 $\times$, (4) 5–6 $\times$, (5) 7–10 $\times$. Values given are the nearest whole number on the enrichment scale. Ranges are given when variation was observed. The sample number (*n*) is the number of insects examined. Unless described as neonate, the larvae were in the last-instar (feeding stage larvae, prepupae or their exuvia cast during pupation). See the materials and methods section for more information about diets. The higher classification of beetles follows that of Lawrence and Newton (1995).

mandibles. These data provided confirmatory evidence that localized areas of larval mandibles were enriched with manganese in *C. angustus* but not in *T. castaneum*.

We investigated the influence of dietary supplementation on the enrichment of metals in the mandibles of *R. dominica*. These data suggested that dietary supplementation may influence the deposition of manganese in mandibles by either influencing whether deposition occurs at all or by influencing the amount of deposition (Table 1). The level of enrichment with manganese and zinc appeared to be higher when the dietary levels were elevated, but more observations would be required to substantiate that trend.

In the dietary supplementation experiment, we examined the exuvium that had been shed at pupation, since preliminary observations indicated that zinc and manganese were not resorbed from larval mandibles during pupation (data not shown). This allowed us to examine the mandibles from both the larval and adult stages of each individual. Zinc was nearly always present in both stages, although manganese was seldom present in both stages (Table 2). When six *R. dominica* were reared on an unsupplemented wheat germ diet, three individuals had manganese in larval but not adult mandibles, two exhibited manganese in adult but not larval mandibles, and one had no manganese in the mandibles of either stage. No individual had enrichment in both stages when reared on the unsupplemented wheat germ diet, and this was also true for the individuals reared on a corn meal diet. All three individuals that were fed a wheat germ diet supplemented with manganese had enrichment in both stages. The numbers of individuals with (3) and without (0) enrichment in both stages was significantly different from the numbers with (0) and without (6) on unsupplemented wheat germ, according to the Fisher exact test. These data indicated that the amount of dietary manganese influenced whether this element was deposited in the mandibles.

After supplementation of the diet with zinc, the majority of the individuals had manganese in both stages, but this was not significantly different from the results obtained on unsupplemented diets (Table 2). About 16% less zinc was added as a supplement, however, when the amounts of the supplements were calculated on a mole basis. Additional observations are needed to show whether zinc can influence the availability of manganese in the insect's diet.

4. Discussion

The mandibles of insects may contain zinc and manganese at concentrations of several percent of the dry weight (Hillerton and Vincent, 1982; Fawke et al., 1997; Quicke et al., 1998). These elements might be present as cations bonded to inorganic anions, proteins, or diphenols. In *Manduca sexta* (L.), the tobacco hornworm moth, the level of diphenols (31%) is about as high as that of protein (35%) and chitin (26%) in the mandibular cuticle (Kramer et al., 1988). Monomeric diphenols are most abundant in the cutting edges of the hornworm mandibles where

Table 2
The effect of dietary supplements of manganese or zinc on the presence of manganese in the mandibles of the lesser grain borer, *Rhyzopertha dominica*[a]

	Proportion of individuals with manganese		
	Larval stage	Adult stage	Both stages
Corn meal	0.4	0.2	0.0
Wheat germ	0.5	0.3	0.0
Wheat germ + Zn	1.0	0.7	0.7
Wheat germ + Mn	1.0	1.0	1.0[b]

[a] The data represent the proportion of individuals that had manganese in the mandibles of their last larval stage, adult stage, or both of these stages. An individual was recorded as not having manganese in both stages if it lacked manganese in one of the stages. Six individuals were examined on each unsupplemented diet, and three were examined on each supplemented diet. See the Materials and methods section for information about the diets.

[b] The numbers of individuals with and without manganese were significantly different from that on either of the unsupplemented diets, according to the Fisher exact test ($P < 0.05$).

zinc also is present (Hillerton and Vincent, 1982; Hopkins et al., 1984; Morgan et al., unpublished data). High concentrations of zinc together with very high concentrations of monomeric diphenols, oxidized diphenols and covalently modified diphenols probably contribute to the hardness of the mandibles.

High levels of zinc are present in the mandibles of newly hatched larvae of the Angoumois grain moth. These larvae must bore through the seed coat to gain access to the nutrients stored in the seed. Zinc is also abundant in mandibles of 11 species of Lepidoptera that feed on moist plant tissues (Hillerton and Vincent, 1982; Fontaine et al., 1991), and their mandibular zinc may contribute to the ability of these larvae to exploit a variety of foods. The adults of these lepidopterans have no mandibles and, therefore, no need to deposit zinc in such structures.

Coleopterans have mandibles in the adult and the larval stages, and might experience budgetary constraints when allocating metals to mandibles. This hypothesis is not supported, however, by the data for the allocation of zinc to the mandibles of different stages. At least five species have zinc in the mandibles of both larvae and adults. One of these species, *C. maculatus*, has mandibles enriched with zinc in several stages of its development, including the first instar larva, the last instar larva, and the adult.

At least two species of seed-eating beetles acquire manganese with an efficiency (5–18%) that is lower than the efficiency (86–94%) with which they acquire zinc (Ernst, 1992, 1993). Therefore, perhaps it is not surprising that at least six species of seed-eating beetles have enrichment with manganese in the mandibles of the adults but not the larvae. In addition to the yellow mealworm, *T. molitor* (Hillerton and Vincent, 1982), these species include *L. serricorne*, *L. oryzae*, *T. castaneum*, *C. ferrugineus*, and *S. granarius*.

The curculionid, *S. granarius*, is an interesting example for study in that this species hatches inside the seed and completes its development within that seed. The absence of both zinc and manganese in larval mandibles of this species suggests that metal hardening is not required for feeding when the outer seed coat has already been penetrated. The adults have manganese in their mandibles, and these structures are located at the tip of a narrow rostrum, which allows the female to chew a hole into the seed, which she may then use as an oviposition site. The rostrum, and its hard mandibles, may have allowed curculionids to exploit seeds as a food for their larvae (Anderson, 1995).

The Bostrichoidea and Cleroidea superfamilies include several primary pests of stored products (*R. dominica*, *L. serricorne*, *S. paniceum*, *G. aequinoctiale*, *T. variabile*, and *T. mauritanicus*). The only one of these pests that lacks zinc in its larval mandibles is the dermestid, *T. variabile*, which instead has high levels of manganese. Neither zinc nor manganese was detected in the larval and adult mandibles of another dermestid, *A. verbasci*. The adults of this species feed on pollen or nectar, whereas the larvae often infest animal products and occasionally wheat flour. The larval mandibles of an anobiid, *G. aequinoctiale*, contain both zinc and manganese. This species, which was originally misidentified in the literature as *G. psylloides* (Czenpinski), develops equally well on wheat kernels or flour (Howe and Burgess, 1952; Belles and Halstead, 1985).

Four of the species that we examined from the Tenebrionoidea and Cucujoidea usually lack zinc and manganese in the larval mandibles (*T. castaneum*, *L. oryzae*, *O. surinamensis*, and *C. ferrugineus*). Stored product beetles from these superfamilies are often categorized as secondary pests that feed on previously damaged kernels, although some of them are difficult to categorize because of their borderline abilities to chew through the seed coat (Sinha and Watters, 1985; Hill, 1990). The last instar larvae of *T. castaneum* are occasionally able to penetrate kernels (White, 1982; Li and Arbogast, 1991). However, even the first instar larvae of *C. angustus* can penetrate intact kernels (Krall and Decker, 1946), and nearly all of the larvae of this species had manganese in their mandibles. The presence of manganese in the mandibles of this species may enhance its ability to chew through the seed coat. The tenebrionids (*T. molitor*, *T. castaneum*, and *L. oryzae*) that lack manganese in their larval mandibles are tenebrionine tenebrionids, while *C. angustus* is a diaperine tenebrionid.

Hillerton et al. (1984) reported the occurrence of zinc or manganese in the adult mandibles of 54 species of stored product beetles. Their results concur with ours regarding the presence of zinc in adult mandibles of *L. serricorne*, *S. paniceum*, and *G. aequinoctiale*, and also the presence of manganese in adult mandibles of *T. variabile*, *O. surinamensis*, *C. ferrugineus*, *T. castaneum*, and *L. oryzae*, and with respect to the absence of metals in adult mandibles of *A. verbasci*. Our results on mandibular metals are the first reported for six species of stored product insects (*S. granarius*, *R. dominica*, *T. mauritanicus*, *C. angustus*, *C. maculatus*, and *S. cerealella*).

Many species of stored product insects are found in packaged foods (Highland, 1991; Mullen, 1994). The larvae of *L. serricorne* and *T. mauritanicus* can bore through many types of packages, whereas those of *O. surinamensis* and *T. castaneum* cannot (Kvenberg, 1974). The mature larvae of *T. mauritanicus* and *T. variabile* can chew through almost all types of packages (Cline, 1978), and the mature larvae of *L. serricorne* can bore through many types. The metal hardening of the mandibles of *T. mauritanicus*, *T. variabile*, and *L. serricorne* may enable these species to penetrate packages.

Some species of Bostrichoidea and Cleroidea, such as *R. dominica*, *L. serricorne*, and *T. mauritanicus*, can utilize wood as a food or as a site for pupation (Fisher, 1950; Lefkovitch, 1967; Hill, 1990). Zinc is present in the mandibles of the larvae of these species. The presence of zinc in mandibles and ovipositors that are used to penetrate wood has been noted in previous studies (Hillerton et al., 1984; Vincent and King, 1995; Quicke et al., 1998). The stored product species in the Tenebrionoidea and Cucujoidea have little or no tendency to bore into wood, although some species (*T. castaneum*, *L. oryzae* and *C. angustus*) occur beneath the bark of dead trees (Sokoloff, 1974). The mandibles of the larvae are either not enriched with any metal or are enriched with modest amounts of manganese.

When *R. dominica* was reared on wheat germ, it usually had zinc in its mandibles but not manganese. Addition of 300-ppm manganese to the wheat germ diet increased the number of insects that deposited manganese in both their larval and adult mandibles. Wheat germ has relatively high concentrations of both manganese and zinc (approximately 100 ppm, Souci et al., 1989) but also has high concentrations of phytic acid, which can limit the absorption of manganese and zinc (O'Dell et al., 1972; Davidson et al., 1995). The binding sites of the phytic acid might become saturated by addition of manganese or zinc ions to the diet, thereby increasing the availability of manganese and zinc. Although the majority of the insects deposited manganese in their mandibles when their diet was supplemented with zinc, further work is needed to determine whether zinc influences the availability of manganese. Our results show that deposition of manganese in the mandibles of *R. dominica* is affected by the amount of manganese in the diet.

Enrichment with manganese can occur in both the larvae and adults of five species (*C. angustus*, *T. variabile*, *S. paniceum*, *G. aequinoctiale*, and *R. dominica*), but four of these species have relatively modest levels of enrichment. Enrichment with zinc occurs in the larvae and adults of most stored product beetles that bore into seeds, packages, or wood. Although enrichment with zinc appears to an important characteristic of these pests, it remains unclear whether zinc may be the most suitable element for hardening mandibles or whether it may simply be a more expendable element than manganese.

Metals are present in mandibles of insect larvae that can bore into seeds, and the insect larvae that do not have metals in their mandibles are usually unable to penetrate seeds. The soil in some regions is deficient in zinc or manganese even to the point of affecting human health (e.g. Cakmak et al., 1999). Crops grown in these regions might have such low levels of these metals that the pest status of the insects that feed upon them could be influenced. Even when feeding on grain with normal amounts of metals, some pests may be unable to acquire enough of the metals for use in the mandibles of all their feeding stages.

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