Toxicity and Repellency of Chenopodium Oil To Four Species of Stored-Product Insects[1,2]

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ABSTRACT Chenopodium oil, an essential oil of *Chenopodium ambrosioides* L., was evaluated for its toxicity and repellency to adults of *Callosobruchus machulatus* (F.), *Sitophilus oryzae* (L.), *Lasioderma serricorne* (F.), and *Tribolium confusum* Jacquelin du Val. Topically applied, the oil was highly toxic to *C. maculatus* (100% mortality at 40 µg/insect) and *L. serricorne* (92.5% mortality at 50 µg/insect), moderately toxic to *S. oryzae* (52.5% mortality at 50 µg/insect), and only slightly toxic to *T. confusum* at 50 µg/insect. When applied to the surface of wheat, the oil was highly repellent to *S. oryzae.* When applied to wheat or black-eyed peas, the oil reduced infestations of *S. oryzae* and *C. maculatus* at dosages of 2000 and 1000 ppm, respectively.

KEY WORDS *Chenopodium ambrosioides,* biological effect, *Callosobruchus maculatus; Sitophilus oryzae; Lasioderma serricorne; Tribolium confusum;* grain protectant, Chenopodium oil.

Chenopodium ambrosioides L. is a strongly aromatic hairy annual or perennial herb, native to tropical America. In the Congo, West Central Africa, *C. ambrosioides* is used by farmers to protect stored beans against the bean weevil, *Acanthoscelides obtectus* (Say), and the cowpea weevil, *Callosobruchus maculatus* (F.), and also to protect stored groundnuts (peanuts) against the groundnut bruchid, *Caryedon serratus* (01.), but according to the farmers, the efficiency is extremely low when the whole plant is used (Delobel and Malonga 1987). Delobel and Malonga also showed that dry powdered *C. ambrosioides* mixed with grain at the rate of 1:40 (w/w) prevented oviposition and produced 90% mortality of *C. serratus* adults.

Chenopodium oil, also called American wormseed oil, is obtained by steam distillation of the above-ground part of the fresh flowering and fruiting plant of *C. ambrosioides* (Guenther 1952). It is used as a fragrance in soap, detergent, lotions, perfumes, and other cosmetic products as a maximum concentration of 0.4%. It also possesses anthelmintic properties in humans (Merck Index 1968), but is now seldom used in pharmaceutical preparations because it has been replaced by synthetic anthelmintics. In addition, it has been shown to be toxic and repellent to insects. For example, at a concentration of 25 ppm, the oil killed 90 - 100% of larvae of the mosquitos *Culex quinquefasciatus* Say (Hartzell and Wilcoxon 1941).

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[2] This paper reports the results of research only. Mention of a proprietary product does not constitute a recommendation or an endorsement by the USDA.

An emulsion dip of the oil killed Japanese beetle larvae, *Popillia japonica* Newman (Leach and Johnson 1925), and the chenopodium (wormseed) powder acted as a short term (about 2 days) repellent to screwworm fly, *Cochliomyia macellaria* (F.) (Parman et al. 1927).

This experiment was designed to investigate the toxicity of chenopodium oil to four species of stored-product insects, and to assess the potential of utilizing low concentrations of chenopodium oil as an insect control agent.

Materials and Methods

The chenopodium oil used in this study was purchased from LaPine Scientific Company, Chicago, IL. Insects used were adults of the cowpea weevil, *C. maculatus* (<5 h old), rice weevil, *Sitophilus oryzae* (L.) (24 - 48 h old), cigarette beetle, *Lasioderma serricorne* (F.) (<72 h old), and confused flour beetle, *Tribolium confusum* Jacquelin du Val (7 - 14 days old). Insects were reared by standard methods and rearing media at 27 ± 1°C and 60 ± 5% RH at the Stored-Product Insects Research and Development Laboratory, Savannah, GA.

For direct contact toxicity studies, a stock solution of 100 mg/ml was obtained by dissolving 100 mg of chenopodium oil in acetone (reagent grade) for a total volume of 1 ml. Lower concentrations of 80, 60, 40, and 20 mg/ml were obtained by dilution of the stock solution with acetone. For treatment, insects were anesthetized briefly with CO_2 and then picked up individually with a vacuum needle. Using a microapplicator, 0.5 µl of the solution was applied to the dorsum of the thorax of each insect. Twenty adults of each species (unsexed except 10♂ and 10♀ for cowpea weevils) were treated with each dose. After treatment, the insects were placed in 100-mm dia. petri dishes, 10 insects/dish (10 kernels of undamaged soft winter wheat were placed in each dish together with rice weevils), and kept in a room maintained at 27 ± 1°C and 60 ± 5% RH under alternating 12-h light and dark cycles. The insects were examined daily for one week. Those that did not move or respond to the gentle touch of a small probe were considered dead. For controls, the insects were treated with acetone only.

For repellency studies, required amounts of chenopodium oil were dissolved in 1 to 2 ml of acetone and mixed with wheat at concentrations of 2000, 1000, and 500 ppm (w/w) by shaking gently for 2-3 min, then spread over aluminum foil for an hour with occasional stirring. Acetone treated wheat was used as control. These treated samples were evaluated for repellency by using the modified Loschiavo food preference apparatus as described by Laudani and Swank (1954). The apparatus consisted of a circular platform 50 cm in dia. with a 50 cm metal rim. The platform had 12 holes, 8.75 cm in dia., equally spaced along the outer edge to accommodate paper cups which were filled to the rim (about 60 g each) even level to the platform with oil treated or acetone treated grain samples. The center of the wheel-cover had an opening, 1.25 cm in dia. fitted with a plastic tube through which 120 adult rice weevils were introduced. For each experiment, three cups were filled with acetone treated wheat, and three each were filled with wheat whose surface was treated with the oil at each of the dosages. The cups were arranged in a different, repeated sequence for each experiment (A-B-C-D- for test 1, A-C-B-D- for test 2, A-B-D-C- for test 3). After a 24 h exposure period in a room at 27 ± 1°C and 60 ± 5% RH, the insects in each cup were collected and counted to determine their distribution. The experiment was repeated three times.

For the protectant studies, wheat or black-eyed peas were surface-treated at dosages of 2000, 1000, and 500 ppm (w/w). Required amounts of chenopodium oil were dissolved in 2-3 ml of acetone and applied to wheat or black-eyed peas as described previously. The treated samples were air-dried and kept in glass jars overnight in a room maintained at 27 ± 1°C and 60 ± 5% RH before use. Acetone-treated wheat or acetone-treated black-eyed peas were used as controls.

For the exposures, a plastic ring (90 mm dia. × 5 mm high) was placed in the center of a glass crystallizing dish (160 mm dia. × 100 mm high), and a 20-g lot of test sample was placed inside the ring which acted as a barrier to prevent the sample from scattering. Ten pairs (10♂ and 10♀) of adult rice weevils (for wheat) or five pairs (5♂ and 5♀) of adult cowpea weevils (for black-eyed peas) were placed on each sample. The dish was covered with a plexiglass lid which had two 35 mm dia. holes covered with 40-mesh metallic cloth, opposite to each other, 20 mm from the edge. The dishes were kept in a room at 27 ± 1°C and 60 ± 5% RH. After seven days of exposure, the insects inside and outside the ring were removed and examined for mortality. Then each sample was kept in a 0.237-liter glass jar and stored in the exposure room. Four weeks after exposure, all samples were examined every 2 or 3 days for F_1 emergence. When emergence started, the insects were counted every 2 or 3 days and removed. The readings were stopped 8 weeks after exposure for rice weevils and 7 weeks after exposure for cowpea weevils in order to avoid including the F_2 insects. The number of F_1 adults was used as a criterion for evaluation. The comparison of F_1 emergence in the treated sample to that in the contol is an index of effectiveness of the treated material in preventing infestation. Only the experiments of same date and with the same insect colony were used in the comparison.

Data were analyzed by using analysis of variance and Duncan's multiple range test (Duncan 1955).

Results and Discussion

Topically applied, chenopodium oil was highly toxic to cowpea weevil and cigarette beetle, moderately toxic to rice weevil, and only slightly toxic (almost nontoxic) to confused flour beetle at the test dosages up to 50 μg/insect. The oil killed all cowpea weevil at 40 μg/insect and caused 92.5% mortality in cigarette beetle at 50 μg/insect (Table 1). It produced 52.5% mortality with rice weevil at 50 μg/insect, but only 15% mortality to confused flour beetle at this dosage.

Chenopodium oil was shown to be a strong repellent to rice weevil (Table 2). The treatments resulted in distributions of rice weevils of 0.8, 6.7, and 15.3% at 2000, 1000, and 500 ppm, respectively, compared with 76.9% in the control. Even at the lowest dosage of 500 ppm, the percentage of insects present was still only 1/5th that of the control.

The protection of wheat and black-eyed peas was evaluated by surface treatment of the commodities with chenopodium oil and then exposure of treated samples to rice weevils and cowpea weevils, respectively. At a dosage of 2000 ppm, the oil was toxic to both species of insects, being more toxic to rice weevil than to cowpea weevil (Table 3). Oil remained toxic, although slightly, to rice weevil at the dosage

Table 1. Direct contact toxicity of chenopodium oil to four species of adult stored-product insects (cowpea weevil - CW; rice weevil - RW; cigarette beetle - CB; and confused flour beetle - CFB).

Dose	Avg. % mortality* ± SEM† at 72 h after application			
(µg/insect)	CW	RW	CB	CFB
50		52.5 ± 1.1 ab	92.5 ± 4.8 a	15.0 ± 6.5 a
40	100.0 ± 0 a	55.0 ± 6.5 a	50.0 ± 12.9 b	5.0 ± 2.0 ab
30	57.5 ± 8.5 b	30.0 ± 7.1 b	42.5 ± 12.5 b	5.0 ± 2.9 ab
20	32.5 ± 17.9 b	7.5 ± 4.8 c	25.0 ± 8.7 bc	2.5 ± 2.5 b
10	0 c	2.5 ± 2.5 c	7.5 ± 7.5 c	2.5 ± 2.5 b
Acetone only	0	0	0	0

* Average of 4 replicates; 10 insects per replicate.

† SEM = Standard error of the mean. Values followed by the same letter within a column per species of insect are not significantly different at the 0.05 level according to Duncan's multiple range test.

Table 2. Distribution of rice weevil adults in untreaed wheat and wheat treated with chenopodium oil following a 24 h exposure in a repellency wheel.

Dose (ppm)	Avg. distribution (%) of insects ± SEM*,†
2000	0.8 ± 0.5 a
1000	6.7 ± 1.9 a
500	15.3 ± 3.2 b
Acetone only	76.9 ± 1.5 c

* Distribution of 120 insects with three samples of each treatment in each experiment, averge of three experiments. SEM = standard error of the mean.

† Values followed by the same letter within a column are not significantly different at the 0.5 level according to Duncan's multiple range test.

of 1000 ppm, but was nontoxic to cowpea weevil. Neither insect species was affected upon exposure to the 500 ppm treatment.

When the number of F_1 adults was used to evaluate the effectiveness of the oil as a protectant for the commodities, a reduction of 83.6% in F_1 progency of rice weevils was obtained at the 2000 ppm of oil on wheat, 24.9% reduction at 1000 ppm, but only 7.2% reduction at 500 ppm. The latter was not significantly different from that of the untreated control.

For the black-eyed peas treated at 2000 ppm, only one of the three samples resulted in F_1 emergence of cowpea weevil; oil resulted in 98.8% reduction of F_1 progency when compared with the contol. At 1000 ppm, the reduction of F_1 progency was 73.2%, and only 7.4% in the 500 ppm treatment.

Results of these studies show that chenopodium oil is toxic to cowpea weevil, rice weevil, and cigarette beetle, and is repellent to rice weevil. Also it protects wheat against infestation by rice weevil and black-eyed peas against infestation by cowpea weevil. Chenopodium oil has significant potential as an insect control agent, and therefore, further investigations with chenopodium oil seem warranted.

Table 3. Toxicity of chenopodium oil-treated wheat to adult rice weevils and of treated black-eyed peas to adult cowpea weevils.

Dose	% Mortality after 7 d		Avg. no. F_1	
(ppm)	M	F	emerg. ± SEM*, †	% Reduction
		Rice Weevils		
2000	86.7	86.7	29.5 ± 13.1 a	83.6
1000	13.3	10.0	135.3 ± 13.6 b	24.9
500	0	0	167.2 ± 2.7 c	7.2
Acetone only	0	0	180.2 ± 6.1 c	0
		Cowpea Weevils		
2000	66.7	13.3	3.3 ± 3.3 a	98.8
1000	0	0	71.3 ± 9.4 b	73.2
500	0	0	246.7 ± 21.8 c	7.4
Acetone only	0	0	266.3 ± 22.5 c	0

* Ten pairs (10♂ and 10♀) of rice weevils in each wheat sample and five pairs (5♂ and 5♀) of cowpea weevils in each black-eyed pea sample; three replicates in each test dose.

† SEM = Standard error of the mean. Values followed by the same letter within a column are not significantly different at the 0.5 level according to Duncan's multiple range test.

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