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Fumigant Activity of Essential Oil from *Armoracia rusticana* (L.) against *Plodia interpunctella* (Lepidoptera: Pyralidae)

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Abstract: The fumigant toxicity of essential oil from Horseradish plant, *Armoracia rusticana* (L.) was assessed against the Indian meal moth, *Plodia interpunctella* (Hübner). Eggs, larvae, pupae and adults were exposed to different concentrations of 0, 2, 4, 8, 16 and 32 μL/L of essential oil. *A. rusticana* oil was found to significantly affect the egg hatch rate, pupal survival, larval and adult mortality of *P. interpunctella*. Results showed that the LC_{50} value for larvae was 15.53g/m^3, as well as 5.54g/m^3 for adults. Fumigation activity of the *A. rusticana* oil appeared to be dose dependent. An increase in treatment concentration led to reduced hatch rate in eggs, and an increase in mortalities of both larvae and adults. Although there was significant difference in survival between the pupae that received plant oil fumigation and those in control treatments, but it appears that pupal eclosion was generally high across treatments, since a high percentage (69%) of pupae eclosed as compared to 90% eclosion observed in the control. These results indicate that it may be possible to achieve toxicity levels similar to those of standard chemical fumigants through the applications essential oils from *A. rusticana*.

Key words: plant essential oil, *Plodia interpunctella*, fumigant activity

Introduction

Insect pests cause a great deal of losses of stored food products. The Indian meal moth, *Plodia interpunctella* (Hübner), is one of the major lepidopteran pests of stored products in China and around the world, causing serious losses in stored produce [1,2,3]. The complete development of this moth takes 27 days from egg to adult at optimal temperature of 30 ℃, 70% r. h. and controlled photoperiod of 16 h light and 8 h dark [4]. Agrochemicals are frequently being used to counter the attack by insect pests. However, these chemicals are faced with great criticisms for their non-environmentally friendly effects and high costs. The foregoing demerits of agrochemicals have lead to the quest for alternative control measure.

Horseradish, *Armoracia rusticana* (Linn.) is a perennial plant of the Brassicaceae family, which includes mustard and cabbages. The plant is probably native to southeastern Europe and western Asia, but is popular around the world today. It grows up to 1.5 metres (five feet) tall and is mainly cultivated for its large white, tapering root[5].

The root is the only part now used, and in the fresh state only. It is nearly cylindrical, except at the crown, where it is somewhat enlarged. It contains potassium, calcium, magnesium and phosphorus, the chief produce being the volatile oil called allylisothiocyanate (AITC), which is identical with that of Black Mustar, which is an antibiotic: protecting food against pathogens. This volatile oil, which is easily developed by scraping the root when in a fresh state, does not pre – exist in the root, the reaction not taking place in the root under normal conditions, because the Sinigrin and Myrosin exist in separate cells, and it is only the bruising of the cells that brings their contents together [6].

The oil is highly diffusible and pungent on account of the Myrosin contained, 1 drop being sufficient to odorize the atmosphere of a whole room. On exposure to the air, the root quickly turns colour and loses its volatile strength[7].

Materials and Methods

Insect Culture and Source

Plodia interpunctella (Hübner) used in this study were obtained from laboratory colony

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maintained on artificial diet consisting of cracked wheat (1000 g), wheat shorts (1000 g), wheat germ (100 g), brewer's yeast (80 g), sorbic acid (4 g), methyl – *p* – hydroxybenzoate (4 g), glycerine (240 mL), pure honey (240 mL) and 120 ml of water[8]. The insect was reared at 29 (±1)℃, 40 60% relative humidity (r. h.), with a 14:10 h light: dark regime.

Source of *Armoracia rusticana* and Extraction of Essential Oil

A. rusticana used in this investigation was obtained fresh during May 2007 from the Teaching and Research farm of Huazhong Agricultural University, Wuhan Hubei China.

Fumigation Bioassay

Fumigation bioassays were carried out in 500ml glass conical flask (fumigation chamber). Essential oil was applied onto a filter paper (1cm 4cm) and suspended vertically within the chamber by thread, together with insect cage. The fumigation chamber was covered with a rubber stopper and sealed with an adhesive tape. Twenty adult *P. interpunctella* (0 – 2 d old), were placed in the cylindrical cage (9cm 3cm), which was perforated with small holes to allow the penetration of the gas. *A. rusticana* oil was applied at the dose of 0 (control), 2, 4, 8, 16 and 32 g/m^3 and each treatment had 3 replicates. Controls received filter paper alone. Percentage insect mortality was recorded after 72 h of exposure to the essential oil gas.

Fumigation of Eggs with the Essential Oil of *A. rusticana*

Black cloth (12cm × 12cm) was placed in a jar, then 15 pairs of newly emerged (0 – 24 h old) adult of *P. interpunctella* were introduced into the jar and were allowed to oviposit on the cloth for 24 h. Afterwards, the cloth bearing the freshly laid eggs was removed and counted. Black cloth bearing 30 eggs was placed in a 500ml conical flask. The essential oil was introduced at different dose rates as described in the previous section. After fumigation, the gas was released from the chamber and eggs were held for 5 days at 29 ± 1℃, until mortality could be determined by the presence or absence of hatching in both treated and untreated (control) eggs.

Fumigation of Larva with the Essential oil of *A. rusticana*

Newly laid eggs were placed in glass together with small quantity of artificial diet, the bottle was covered with organdy screen and incubated at 29(±1)℃. The resulting 3rd instar larvae emerging 2 d after hatching were selected for this experiment. It should be noted that result from a preliminary culture showed that 3rd stadium of *P. interpunctella* were obtained 12 d after hatching. Thirty *P. interpunctella* larvae (3rd instar) were selected from the stock insect culture and placed in a cylindrical insect cage (4.5cm × 1.5cm), together with small amount of artificial diet (5g), then capped with rubber stopper, the larva were used for the bioassays. Thereafter, the essential oil together with the insect cage was introduced into the arena as described in the previous section. After fumigation, the gas was released and the larvae was held for 5 days at 29 ± 1℃, then larval mortality was counted in both treated and control arenas.

Fumigation of Pupa with the Essential Oil of *A. rusticana*

Thirty pupae (30) of *P. interpunctella* were placed in a perforated insect cage (4.5cm × 1.5cm), capped with rubber stopper, the cage and the essential oil were suspended into the fumigation chamber as earlier described. After fumigation, the gas was released from the chamber and held for 7 days at 29 ± 1℃, until mortality could be determined by the presence or absence of adult emergence in both treated and control pupae.

Statistical Analysis

Data from egg hatch, larval, pupal and adult mortality subjected to analysis of variance and where significant differences existed, means were compared using Tukey's b test. Because percentage hatch rate, larval and adult mortality were not normally distributed, data were first normalized by arcsine transformation before analysis. This consists of taking the arcsine of the square root of a number (y = sine (x) ^ – 1). After analysis, data were back – transformed by squaring the sine of the number[9].

Results

Effect of *A. rusticana* Oil on Eggs of *P. interpunctella*

The fumigant effect of differnrent concentrations of *A. rusticana* oil on eggs of *P. interpunctella* is shown in Fig. 1. *A. rusticana* oil affected percentage hatch rates significantly ($P < 0.001$). The fumigant activity of the oil on the eggs was dose dependent. At lowest tested dose (2 μL/L), there was high percentage hatchability and this was not significantly ($P < 0.05$) different from that observed in the control. Whereas significantly lower percentages

hatch rates were observed at higher doses. This is suggestive that *A. rusticana* had fumigant action against the eggs of *P. interpunctella*.

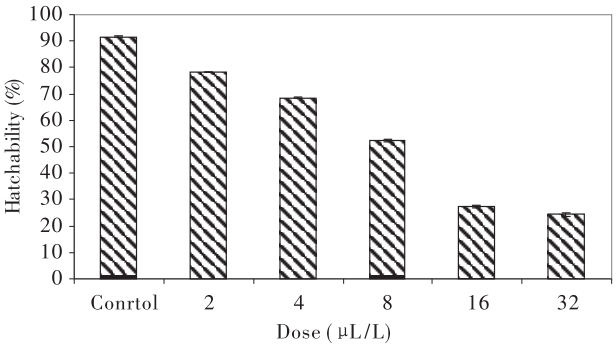

Fig. 1 Effect of fumigation with varying concentrations of *Armoracia rusticana* oil against eggs of *Plodia interpunctella*. Columns followed by the same alphabet(s) are not significantly different ($P < 0.001$) using Tukey's b test.

Effect of *A. rusticana* Oil on Larvae of *P. interpunctella*

The percentage mortality of *P. interpunctella* larvae after exposed to different doses of *A. rusticana* oil is represented in Fig. 2. Fumigation with *A. rusticana* oil had significant effect on larval mortality at all treatment levels in relation to the control ($P < 0.001$). As concentration increases, larval mortality increases. The highest mortality was observed among larvae groups that were exposed to 32 μL/L dose of *A. rusticana* oil.

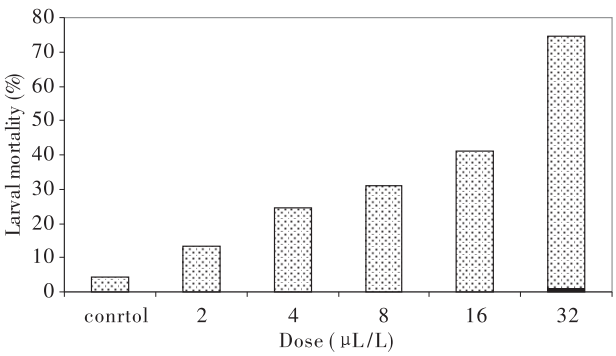

Fig. 2 Fumigant effect of *Armoracia rusticana* oil at different concentrations against larvae of *Plodia interpunctella*. Columns followed by the same alphabet(s) are not significantly different ($P < 0.001$) using Tukey's b test.

Effect of *A. rusticana* Oil on Pupae of *P. interpunctella*

After pupae of *P. interpunctella* were fumigated with oil of *A. rusticana*, there was significant ($P < 0.013$) difference in the number of pupa that became adult (eclosed) Fig. 3. It was generally observed that there was high percentage pupal survival across treatments. However, there was no significant ($P > 0.05$) difference in the percentage survival of among the pupae that were exposed to 2, 4 and 8 μL/L and between those exposed to 16 and 32 μL/L doses of plant oil.

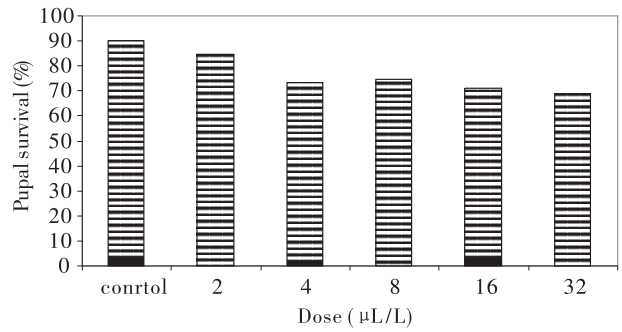

Fig. 3 Fumigant effect of *Armoracia rusticana* oil at different concentrations against pupae of *Plodia interpunctella*. Columns followed by the same alphabet(s) are not significantly different ($P < 0.013$) using Tukey's b test.

Effect of *A. rusticana* Oil on Adults of *P. interpunctella*

Fig. 4 shows the effect of fumigation with varying concentrations of *A. rusticana* oil against adults of *P. interpunctella*. *A. rusticana* oil had significant ($P < 0.001$) effect on *P. interpunctella* adult mortality. The fumigant activity of the oil on the adults was dose dependent, with percentage mortality increasing in relation to increase in treatment concentration. The highest mortality was recorded in insects exposed to 32 μL/L, however, this was not significantly ($P > 0.05$) different from mean percentage mortality of adults exposed to 16 μL/L of plant oil. This indicates that *A. rusticana* had fumigant activity against the adults of *P. interpunctella*.

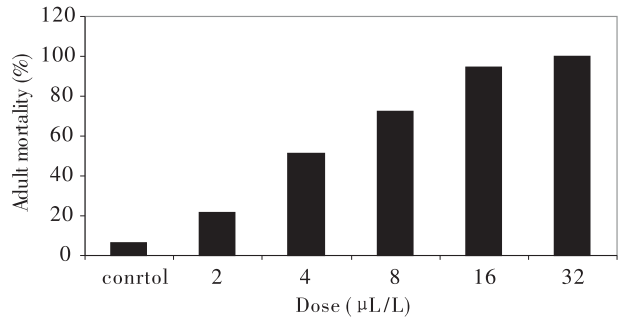

Fig. 4 Effect of fumigation with varying concentrations of *Armoracia rusticana* oil against adults of *Plodia interpunctella*. Columns followed by the same alphabet are not significantly different ($P < 0.001$) using Tukey's b test.

Discussions

The essential oil of *A. rusticana* was

shown here to possess fumigant bioactivity against *P. interpunctella* (adults, pupae, larvae and eggs). It caused high percentage mortality of larvae and adult insects exposed to 32 μL/L gas vapour of *A. rusticana*, and hatchability was also inhibited at this dose. The oil however, appeared not to have much impact on pupal eclosion, since a high percentage (69%) of pupae eclosed as compared to 90% eclosion observed in the control.

The reason for the high fumigant effect of *A. rusticana* oil on eggs, larvae and adults of *P. interpunctella*, could be attributed to its high pungent odour due to the presence of AITC in the volatile oil. It could be that the volatile oil is able to block the spiracles of the insects by impairing breathing and thereby chocking them to death[10,11]. Its relatively low fumigant effect on pupal survival might be that the gas vapour could not permeate through the thick wall of the pupal case.

A number of toxic chemicals produced by plants elicit pungent sensation in mammals [12,13]. The efficiency of the WasaOuro system, an insecticide based on AITC, the active component responsible for insecticidal action of horseradish and other brassicaceae family, was found to possess fumigant action against *Lasioderma serricorne* and *Tribolium confusum* by disrupting normal reproductive cycles of both insects, resulting in an insect population reduction in grain foods[14]. Natural toxin of isothiocyanates including AITC, has been shown to have insecticidal activities. AITC was reported to increase the production of carbon dioxide in the American cockroach [15]. Fumigant activities of horseradish and garlic oils against *Lycoriella ingenua* (Diptera: Sciaridae) have been reported[11].

Increasing problems concerning the use of modem synthetic chemical insecticides, such as persistence of residues, resistance, and damage to the environment and human health have generated interest in naturally occurring products. It should be noted that biologically active compounds of food plants are assumed to be environmentally more acceptable and less hazardous than others to humans. The results presented in this study suggest that *A. rusticana* oil or its major constituents could be efficient fumigants and also could be integrated with other pest management procedures. Further studies are needed to assess the fumigant activity of this essential oil and its constituents to other insects. Also, detailed mechanism of action of AITC in target pests could be an interesting area of research.

Acknowledgements

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