

Repellent effect of whole bean flour against *Sitophilus zeamais* (Motschulsky)

(Coleoptera: Curculionidae)

ผลการขับไล่ของผงถั่วต่อด้วงงวงข้าวโพด

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ABSTRACT

The repellent effect of whole flour of red kidney bean, mung bean and navy beans against *Sitophilus zeamais* was evaluated by cup bioassay technique. *Sitophilus zeamais* moving out of rice grains treated with whole flour of all three beans was significantly different among concentrations after 72 h exposure. The repellency of 20% whole flour of red kidney bean and mung bean was significantly higher than that of the control. The repellent effect of 1, 10 and 20% navy bean was significantly higher than that of the control. At 20%, the repellent effect was significant difference among bean type. The mechanism for repellency needs to be investigated.

บทคัดย่อ

จากการประเมินผลการขับไล่ของผงถั่วที่มีต่อด้วงงวงข้าวโพดด้วยเทคนิค cup bioassay พบว่าที่ 72 ชั่วโมง ด้วงงวงข้าวโพดออกจากข้าวที่ผสมกับผงถั่วทั้งสามชนิด มีความแตกต่างระหว่างความเข้มข้นอย่างมีนัยสำคัญ การขับไล่ของผงถั่วแดงและถั่วเขียวที่ความเข้มข้น 20 เปอร์เซ็นต์มีค่าสูงกว่ากลุ่มควบคุม ผลการขับไล่ของผงถั่วขาวที่ความเข้มข้น 1, 10 และ 20 เปอร์เซ็นต์มีค่าสูงกว่าเมื่อเทียบกับกลุ่มควบคุม และการขับไล่ของถั่วแต่ละชนิดมีความแตกต่างกันอย่างมีนัยสำคัญที่ความเข้มข้น 20 เปอร์เซ็นต์ ซึ่งกลไกของการขับไล่จากถั่วแต่ละชนิดจำเป็นต้องตรวจสอบต่อไป

Key Words: Whole flour, cup bioassay

คำสำคัญ : ผงถั่ว ถั่วทดสอบ

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Introduction

Rice is the most important staple crop in Thailand. It is subjected to attack by a group of insect pests during storage and transport (Kengkapanich, 2003). The maize weevil, *Sitophilus zeamais* (Motschulsky) (Coleoptera: Tenebrionidae) is universally regarded as one of the most destructive primary pest of stored product (Hayashi, et al 2004). The weevils frequently start infests in the field before harvesting and they are carried into the storage where the population builds up rapidly (Appert, 1987, Adedire and Lajide, 2003). Voracious feeding on whole grain by this insect result in contamination and reduction of dry weight, nutrition, marketability (lowering of grade) and seed viability (Semple, et al 1992, Trematera, et al 2007). The agriculturist manner to avoid post harvest losses due to this pest is the use of synthetic insecticides. The undesirable effects of synthetic pesticides on environment and human health have become a major concern and thus given incentive to pursue alternative procedures of pest control to reduce the use of synthetic pesticides (Schmutterer and Ascher, 1986).

Legume seeds comprise a wide range of secondary metabolites with toxic and deterrent effects against insect pests (Harborne, et al 1971, Bell, 1978). An addition of yellow split-peas, *Pisum sativum* L., with wheat resulted in a remarkable reduction of survival and reproduction rate of *Sitophilus oryzae* L. (Coombs, et al 1977, Holloway, 1986).

Dethier, et al (1960) described insect repellents as chemical substances which cause the insect to move away from the source of the substance. Repellents have potential for excluding stored-product pests, and have been used to prevent insect feeding and

oviposition (Fields, et al 2001). At present, there is still a need to find repellents which are effective, persistent and practical to use. This study aimed to determine repellency of whole bean flour of three legume seeds against the maize weevil, *S. zeamais*.

Materials and methods

Mass rearing insect

Sitophilus zeamais was obtained from Postharvest Technology Research and Development Laboratory, Postharvest and Product Processing Research and Development Office, Department of Agriculture, Bangkok. One hundred of unsexed maize weevils, 1 to 2 weeks old, were transferred to 200 g milled rice in 400 ml glass bottle which covered with filter paper. The parent weevils were transferred to new oviposited bottles every three days. They were allowed to lay eggs for one month. The weevils were maintained at $27\pm 2^{\circ}\text{C}$, $75\pm 5\%$ R.H. and under 12:12 h (light: dark) photoperiod.

Preparation of whole bean flour

Three legume seeds, i.e. mung bean, *Vigna radiata* (L.) was obtained from Chai Nat Field Crops Research Center Thailand, navy bean, *Phaseolus vulgaris* and red kidney bean, *Phaseolus vulgaris* L. were purchased from Royal Project Foundation of Thailand. The beans were ground to powder with Fitzmill Comminutor 60 mesh size (Fitzpatrick Elhurst, Illinois 60125, USA) followed by Alpine pinmill 100 mesh size. (Augsburg, Germany)

Repellency test

The cup bioassay technique, as described by Pretheep-Kumar, et al (2004), was used to evaluate the repellent effect of whole bean flour against *S. zeamais*. A quantity of 100 grams of rice was treated with each type of whole bean flour at 0, 0.1, 1, 10 and 20% w/w

basis. The treated rice was placed in a covered perforated cup with the hole size of 2-mm diameter. The holes are large enough to allow insects to pass through. Twenty unsexed adults of the maize weevil, 1 to 2 weeks old, were placed on the rice, at the center of the perforated container. The repellency of the whole bean flour was measured in terms of numbers of weevils moving out of the container, away from the treated rice. They were trapped in a tray below the container. The number of trapped weevils was determined at 72 h exposure. The experiment was repeated three times with different sets of weevils.

Statistical Analysis

Numbers of weevils moving out of the containers of treated and untreated rice were analyzed by Kruskal-Wallis one-way analysis of variance. The multiple comparison tests were conducted after the significance of Kruskal-Wallis analysis (Conover, 1980).

Results and discussion

The repellency of whole flour of red kidney bean against *S. zeamais* was significantly different among concentrations (Table 1). Similar result was observed for mung bean and navy bean.

The multiple comparison tests after Kruskal-Wallis one-way analysis of variance are presented in Table 2-4. The significant difference was calculated from the difference between observe different values and the critical different value at $p=0.05$. The repellent effect of 20% whole flour of red kidney bean against *S. zeamais* was significantly higher that of the control (Table 2). Similar outcome was found for mung bean (Table 3). There was no significant difference among repellency of rice grains treated with various concentrations of whole flour of navy bean

(Table 4). The repellent effect of rice grains treated with 1, 10 and 20% whole flour of navy bean against *S. zeamais* was significantly higher that of the control (Table 5).

The repellency of rice treated with 20% whole flour was significantly different among bean type (Table 6). The repellent effect of whole flour at lower concentration was not significantly different among bean type.

Our results indicated that whole flour of red kidney bean and mung bean at 20% could repel *S. zeamais*. Whole flour of navy bean at 1, 10 and 20 % significantly repel *S. zeamais* when compared with the untreated control. Pretheep, et al (2004) found that 1% whole pea flour caused 15% repellency against *Rhyzopertha dominica*.

The repellency of whole bean flour in this study could be reflected by reduced number of weevils on treated rice grains. The mechanism for the reduction was not identified in this study. Based on our results, the repellent effect of whole bean flour tended to depend on concentration and bean type. The repellent effect may be due to some compounds in seeds such as starch, fiber or protein. For example, Bodnaryk, et al (1999) reported that pea starch is not repellent but the pea protein possesses repellent activity against *T. castaneum*, *S. oryzae* and *R. dominica*. Field, et al (2001) studied repellency effect of pea, *P. sativum* fractions against stored product insects. They found that for the three pea fractions, the rank order of toxicity as well as repellency was protein>fiber>starch. They suggested that the repellency may cause by chemosensory effects of pea protein fractions, either olfactory or gustatory.

Table 1 The median of *S. zeamais* leaving out of rice grains treated with whole flour of red kidney bean, mung bean and navy bean after 72 h exposure: Kruskal-Wallis one-way analysis of variance table.

Bean Type	Median of <i>S. zeamais</i>					χ^2	P-value
	Concentration (%)						
	0	0.1	1	10	20		
Red kidney bean	0	3	5	2	10	10.98	0.027 [*]
Mung bean	0	1	4	3	9	11.01	0.026 [*]
Navy bean	0	0	12	12	15	11.89	0.018 [*]

* Significantly difference at p=0.05

Table 2 The observe different values of *S. zeamais* moving out of rice grains treated with whole flour of red kidney bean after 72 hour exposure.

Concentration (%)	Observe different values			
	Concentration (%)			
	0.1	1	10	20
0	5.00 (NS)	8.50 (NS)	5.17 (NS)	11.33* (Sig.)
0.1	-	3.50 (NS)	0.17 (NS)	6.33 (NS)
1	-	-	3.33 (NS)	2.83 (NS)
10	-	-	-	6.17 (NS)

Critical difference = 10.25

* Significantly difference (observe different value > critical difference at p=0.05)

NS, not significant difference

Table 3 The observe different values of *S. zeamais* moving out of rice grains treated with whole flour of mung bean after 72 hour exposure.

Concentration (%)	Observe different values			
	Concentration (%)			
	0.1	1	10	20
0	4.33 (NS)	8.50 (NS)	6.00 (NS)	11.17* (Sig.)
0.1	-	4.17 (NS)	1.67 (NS)	6.83 (NS)
1	-	-	2.50 (NS)	2.67 (NS)
10	-	-	-	5.17 (NS)

Critical difference = 10.25

* Significantly difference (observe different value > critical difference at p=0.05)

NS, not significant difference

Table 4 The observe different values of *S. zeamais* moving out of rice grains treated with whole flour of navy bean after 72 hour exposure.

Concentration (%)	Observe different values			
	Concentration (%)			
	0.1	1	10	20
0	1.00 (NS)	6.33 (NS)	7.50 (NS)	10.16 (NS)
0.1	-	5.33 (NS)	6.50 (NS)	9.17 (NS)
1	-	-	1.17 (NS)	3.83 (NS)
10	-	-	-	2.67 (NS)

Critical difference = 10.25

* Significantly difference (observe different value > critical difference at p=0.05)

NS, not significant difference

Table 5 Comparison between observe different values of *S. zeamais* moving out of rice grains treated with whole flour of navy bean and untreated control after 72 hour exposure.

Concentration (%)	Observe different values			
	Concentration (%)			
	0.1	1	10	20
0	1.00	6.33*	7.50*	10.16*
	(NS)	(Sig.)	(Sig.)	(Sig.)

Critical difference = 4.68

* Significantly difference (observe different value > critical difference at p=0.05)

NS, not significant difference

Table 6 Comparison median of *S. zeamais* moving out of rice grains treated with whole flour of mung bean, navy bean and red kidney bean at the same concentration.

Concentration (%)	Median of <i>S. zeamais</i>			χ^2	P-value
	Mung bean	Navy bean	Red kidney bean		
0	0	0	0	NaN	NA
0.1	1	0	3	4.43	0.11
1	4	12	5	3.62	0.16
10	3	12	2	3.35	0.19
20	9	15	10	6.06	0.05*

* Significantly difference at p=0.05

NaN = not a number

NA = no available

Conclusions

The whole flour of red kidney bean, mung bean and navy bean shows repellent effect against *S. zeamais* at high concentration. The mechanism for repellency should be study further. Based on this result, whole flour of these three beans could be used in combination with other control methods such as biological control. Prior to its application, negative

effect of whole bean flour on natural enemy and non-target pest should be investigated.

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References

- Adedire, CO. and Lajide, L. 2003. Ability of extracts of ten tropical plant species to protect maize grains against infestation by the maize weevil, *Sitophilus zeamais* during storage. Niger. J. Exp. Biol. 4(2):175-179.
- Appert, J. 1987. The Storage of Food grains and Seeds. The Tropical Agriculturist CTA, Macmillian publishers Ltd. London. 146.
- Bell, EA. 1978. Toxins in seeds. In: Harborne, J.B. (Ed.), Biochemical Aspects of Plant and Animal Coevolution. New York, Academic Press Inc. 143-161.
- Bodnaryk, R., Fields, P., Xie, Y., and Fulcher, K. 1999. Insecticidal factors from field pea. United States of America Patent 5,955,082.
- Conover, WJ. 1980. Practical Nonparametric Statistics. John Wiley & Sons, Inc., Canada.
- Coombs, CW., Billings, CJ., and Porter, JE. 1977. The effect of yellow split-peas (*Pisum sativum* L.) and other pulses on the productivity of certain strains of *Sitophilus oryzae* (L.) (Col. Curculionidae) and the ability of other strains to breed thereon.). J. Stored Prod. Res. 13: 53–58.
- Dethier, VG., Barton-Browne, L., and Smith, CN. 1960. The designation of chemicals in terms of the responses they elicit from insects. J. Econ. Entomol. 53: 134–136.
- Fields, PG., Xie, YS., and Hou, X. 2001. Repellent effect of pea (*Pisum sativum*) fractions against stored- product insects. J. Stored Prod. Res. 37: 359– 370.
- Harborne, JB., Boulter, D., and Turner, BL. 1971. Chemotaxonomy of the Leguminosae. Academic Press, London, UK.
- Hayashi, T., Nakamura, S., Visarathanonth, P., Uraichuen, J., and Kengkarnpanich, R. eds. 2004. Stored Rice Insect Pests and their Natural Enemies in Thailand. 1st ed. Funny Publishing Co. Ltd., Bangkok Thailand.
- Holloway, GJ. 1986. The potency and effect of phytotoxins within yellow-pea (*Pisum sativum*) and adzuki bean (*Vigna angularis*) on survival and reproductive potential of *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae). Bull. Entomol. Res. 76: 287–295.
- Kengkarnpanich, R. 2003. Comparative Study of Phosphine Concentration and Effectiveness in Different Commodities after Fumigation. Master of Science (Agriculture). Kasetsart University.
- Pretheep-kumar, P., Mohan, S., and Ramaraju, K. 2004. Protein-enriched pea flour extract protects stored milled rice against the rice weevil, *Sitophilus oryzae*. J. Insect Sci. 4: 96.
- Schmutterer, H., and Archer, KRS. 1986. Natural pesticides from the neem trees (*Azadirachta indica* A Juss) and other tropical plants. Proc. 3rd Neem Conf. Nairobi. 517-523.
- Semple, RL., Hicks, PA., Lozare, JV., and Castermans, A. 1992. Towards integrated commodity and pest management in grain storage: A Training Manual for application in humid tropical storage systems. Food and Agricultural Organization of the United Nations, Regent.

Trematerra, P., Valente, A., Athanassiou, CG., and

Kavallieratos, NG. 2007. Kernel-kernel

interactions and behavioural responses of the

adults maize weevil *Sitophilus zeamais*

Motschulsky (Coleoptera: Curculionidae).

Appl. Entomol. Zool. 42(1): 129-135.