Repellent effect and ovipositional inhibition of essential oil formulas from star anise (*Illicium verum*) and dill (*Anethum graveolens*) against adult of red flour beetle (*Tribolium castaneum* (Herbst)) and saw-toothed grain beetle (*Oryzaephilus surinamensis* (L.))

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Abstract

Repellent effect and ovipositional inhibition properties of essential oil formulas obtained from star anise (*Illicium verum* Hook.f.) and dill (*Anethum graveolens* Linn.) against adult of red flour beetle (*Tribolium castaneum* (Herbst)) and saw-toothed grain beetle (*Oryzaephilus surinamensis* (L.)) were conducted. The different proportions between star anise: dill as 4:0, 3:1, 2:2, 1:3 and 0:4, presented as formulas S4D0, S3D1, S2D2, S1D3 and S0D4, respectively were used in the experiments. The bioassays were choice test done in repellent tube (2 cm in diameter, 20 cm long) with rice seeds and essential oil formulas at concentrations of 2, 4 and 6%, volume of 50 µl, later recorded at 72 hr, then compared with control (95% ethanol). The ovipositional inhibition caused by each essential oil formula was evaluated by following previous experiment when the emergent adult developed from laid egg of each tube was checked after 45 days. The result showed that S3D1 formula was at 6% concentration was the best performance. It presented %repellent Index against adult of red flour beetle and saw-toothed grain beetle at 41.7% and 32.0%, respectively. In the same way, this formula was also highly effective on ovipositional inhibition presented the number of 35.6% adult red flour beetle which was significantly different from the control. In the other hand, of ovipositional inhibition on saw-toothed grain beetle was no significantly different to the control.

Keywords: repellent index, ovipositional inhibition, essential oil formula

1. Introduction

Stored product pests are major causes of degradation in dried products and grains (Rajendran, 2002). These pests, particularly red flour beetle and saw-toothed grain beetle, are normally small and unnoticeable unless the outbreaks have resulted in extensive damages. Moreover, the life cycles of the pest are relatively short. This results in high populations in a short period of time (Rees, 2004), and consequently produces infestations. In general, evidence of the infestation can be seen in damage such as gnawing, nesting and defecating (Department of Agriculture, 2005). The degradation can be weight loss, nutrition loss and germination loss. Today, stored product pest management usually employs pesticide fumigation to eliminate infestations. In particular, methyl bromide (CH_3Br) and phosphine are among the highly used products. Methyl bromide is popular for its high diffusing ability and infiltration into the products. In addition, this chemical can kill insects in every stage. Nonetheless, methyl bromide is classified as hazardous material (class I) which usually impacts animals, environment, consumers and even the pesticide users. It was also reported as having 60 times higher ozone depletion property than CFCs (WMO, 1995). Phosphine is also considered

dangerous. A high dose application can inhibit germination (Sittisuang and Makakita, 1985). Moreover, continual single applications can lead to resistance of insects. (Pimentel, 2007).

Recently, alternative and environmental friendly approaches have been studied and introduced, for example biological control, crop improvement, vacuum storage, integrated control, and biopesticides. There are over 1,000 medical plants reported as having pest control and repellence properties (Jirathamsri, 1993). The plant extracts are usually applied in different methods including fumigants, contact residues, antifeedants, and repellents. Therefore, plant extracts and essential oils are considered a potential natural alternative in management of stored product pests. Thanasirungkul et al., (2013) reported the biopesticide property of star anise (*Illicium verum* Hook.f.), dill (*Anethum graveolens* Linn.), clove (*Syzygium aromaticum* (L.) Merr. & L.M.Perry) and lemon grass (*Cymbopogon citrates* (Dc.ex.Nees)) against adults of red flour beetle (*Tribolium castaneum* (Herbst)), lesser grain borer (*Rhyzopertha dominica* (Fabricius)) and corn weevil (*Sitophilus zeamais* Motshulsky) with more than 70% mortality. In addition, essential oils from star anise, dill, clove and lemon grass could control saw-tooth grain beetle with LC_{50} at 7.170 µl/L air (Thanasirungkul et al., 2012) Suthisut et al. (2014) reported repellent activity of essential oil from Zingiberaceae plants against maize weevil and red flour beetle in Petri-dish choice test. The results showed that essential oil from turmeric repelled more than 90% of both insects.

It can be seen here that plant essential oils show effective stored product pest management properties. In addition, as they are naturally derived products, essential oils are often harmless to human, animal, and environment, even when used regularly and repeatedly on the same stored product. Plant essential oils have become a potential alternative for the management of stored product pests which have short life cycle and high reproduction. Therefore, this study investigated in vitro repellent effect and ovipositional inhibition of essential oils form star anise and dill on red flour beetle and saw-tooth grain beetle.

2. Material and Methods

2.1. Insect culture

Red flour beetle and saw-tooth grain beetle in rice seed collected from rice mills in Ladkrabang, Bangkok, Thailand were selected and cultured at room temperature in plastic box sized 27x18x10 cm with net covering on the top. The insects were fed with brown jasmine rice. Adults 10-15 days-old of the 2^{nd} – 3^{rd} generations were used for tests.

2.2. Plant essential oil extraction

Dried flowers of star anise and dried seeds of dill were extracted using water distillation method in which the plant were boiled for 6 hr. The suspended oils were then collected and kept in amber bottles at 12°C.

2.3. Essential oil formulas

Essential oils from star anise and dill were mixed at different ratios including

S4D0	=	star anise : dill	ratio	4:0
S3D1	=	star anise : dill	ratio	3:1
S2D2	=	star anise : dill	ratio	2:2
S1D3	=	star anise : dill	ratio	1:3
S0D4	=	star anise : dill	ratio	0:4

2.4. Examination of repellent property

The repellent test in this study was modified from Pumnuan et al., (2012) with 3 replications. Red flour beetle and saw tooth grain beetle (50 each) were transferred into the center of repellent tube sized 2 cm in diameter and 20 cm in length filled with brown jasmine rice. One end of tube was earlier installed with filter paper (Whatman No.1sized 1.8 cm^2 in diameter) dropped with each essential oil formula at the concentration of 2, 4 and 6% (50 µl) and 10 min left dry at room temperature. The other end was control (Ethanol 95%). The results were observed and recorded at 12 hr after the treatments. The obtained data were analyzed for %repellent index with $[(N_c + N_t)/ N_c + N_t)]\times100$ (Pascual-Villalobos and Robledo, 1998) when, N_t = the number of insects found in the treatment and N_c = the number of insects found in the control.

2.5. Examination of ovipositional inhibition property

It was expected that the insect eggs were laid in the rice seeds. Therefore, treated rice seed in the repellent tubes were further reserved for the incubation in plastic boxes sized 6x9.7x3.6 cm. The incubated adults were observed on day 30 after the treatments. The obtained results were subsequently to the number of the adult insects found in the control

3. Results and Discussion

The results of essential oil repellency examination showed that the essential oil formula S3D1 at the concentration of 6% demonstrated the highest quality in repelling red flour beetles with repellent index of 41.7%. The following was essential oil formula S4D0 at the concentration 6% with repellent index of 38.7%. On the other hand the control presented the repellent index of -1.7% (Figure1). The result of essential oil formula repellency examination against saw-tooth gain beetle showed that essential oils formula S3D1 at the concentration of 6% demonstrated the highest repellency with the repellent index of 32.0%, followed by this formula at the concentration of 4% which showed repellent index of 17.7%, while the control showed -1.2% (figure 2).

The examination of ovipositional inhibition showed the essential oil formula S3D1 at the concentration of 4% demonstrated the highest ovipositional inhibition with 35.6% observation of adult red flour beetle. The results were significantly different from the control (50.9%). The following was S3D1 at the concentration of 6% with 38.9% observation of adult red flour beetle (Figure3). Remarkably, the results from all essential oil formula treatments against saw-tooth gain beetle showed no significant differences when comparing to the control. It presented 42.7-61.1% adults appeared in treatments (Figure 4). The result was congruent with Jawsuwanwong et al. (2014) which examined repellent property of the essential oil formula of star anise and dill against corn weevil by choice test in petri-dish and repellent tube. It was found in the study that S3D1 demonstrated 90% repellency in petri-dish choice test and 50% repellent tube choice test at 2-6 hr after the treatment. In addition, 28.1% of adult corn weevils were observed examination of ovipositional inhibition. Moreover, Ho et al. (1995) reported the insecticidal property of star anise essential oil against more than 70% eggs, larvae, and adults of corn weevil.

4. Conclusion

The essential oil formula of star anise and dill at the ratio of 3:1 presented the highest repellency against red flour beetles and saw-tooth grain beetle with the repellent index of 41.7

and 32.0% respectively. In addition, the formula presented the highest ovipositional inhibition in red flour beetle with 35.6% observation of the adult insects.

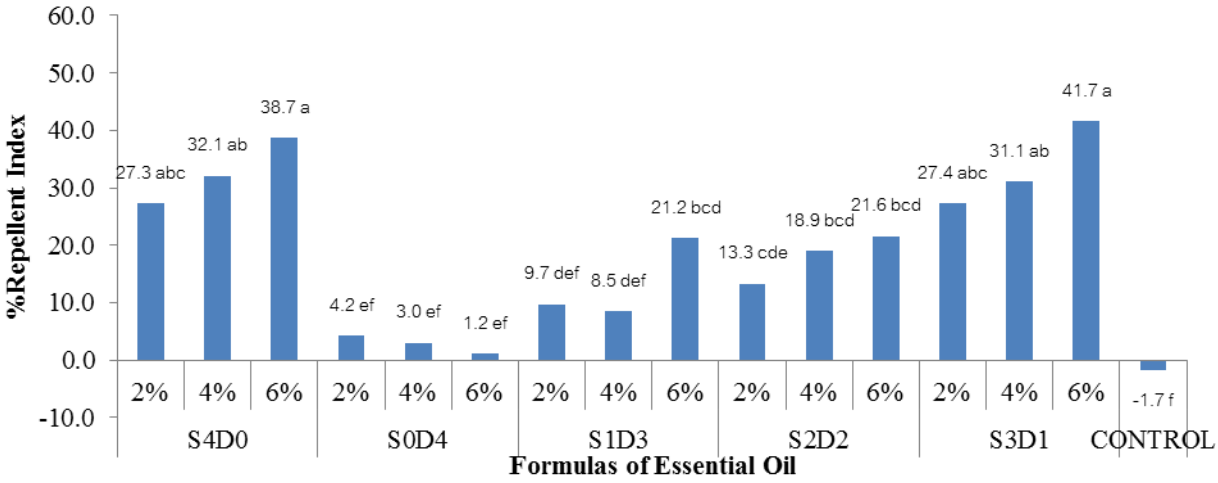

Figure 1 Percentage of repellent index (%RI) of essential oils of medicinal plants against the red flour beetle, *Tribolium castaneum* Herbst at 72 hours by fumigation method.

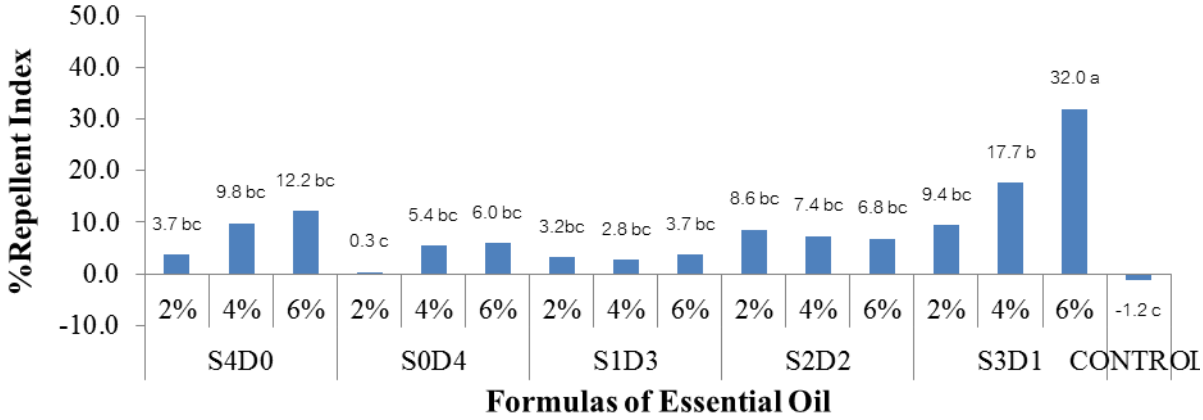

Figure 2 Percentage of repellent index (%RI) of essential oils of medicinal plants against the saw-toothed grain beetle, *Oryzaephilus surinamensis* Linn at 72 hours by fumigation method.

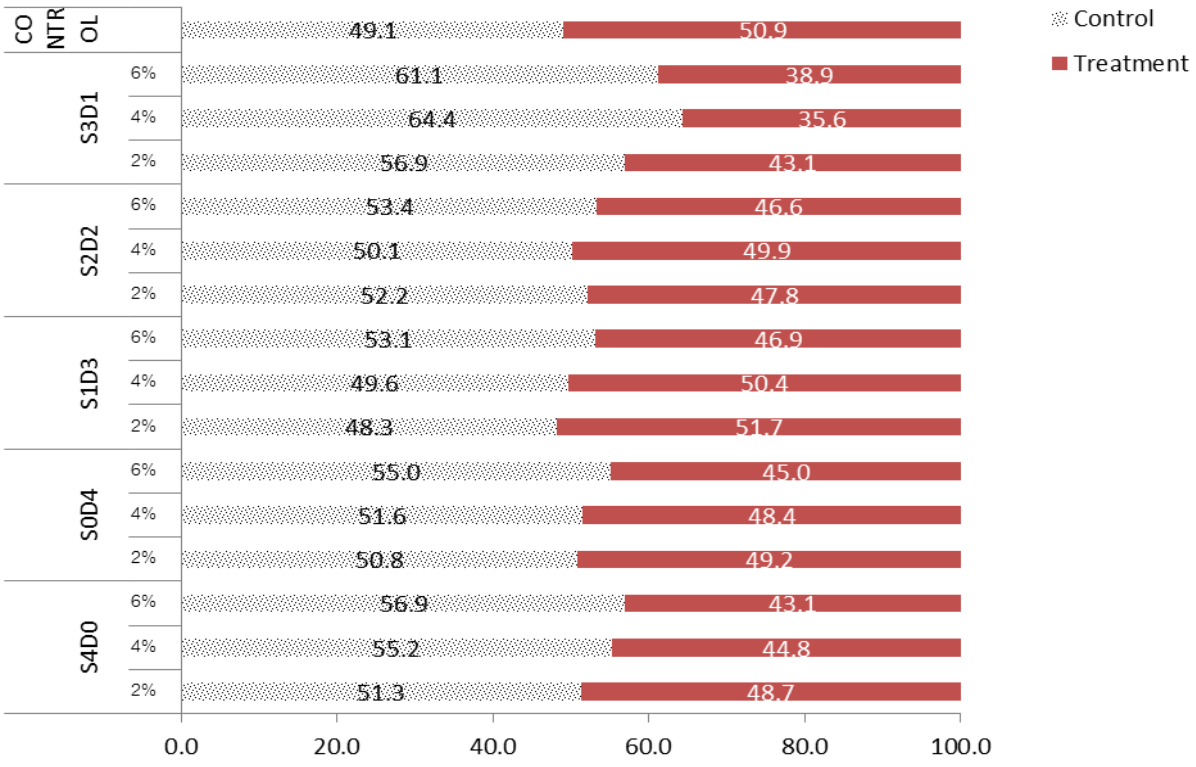

Figure 3 Number of adults of red flour beetle, *Tribolium castaneum* Herbst developed from egg after treated with essential oil formulas.

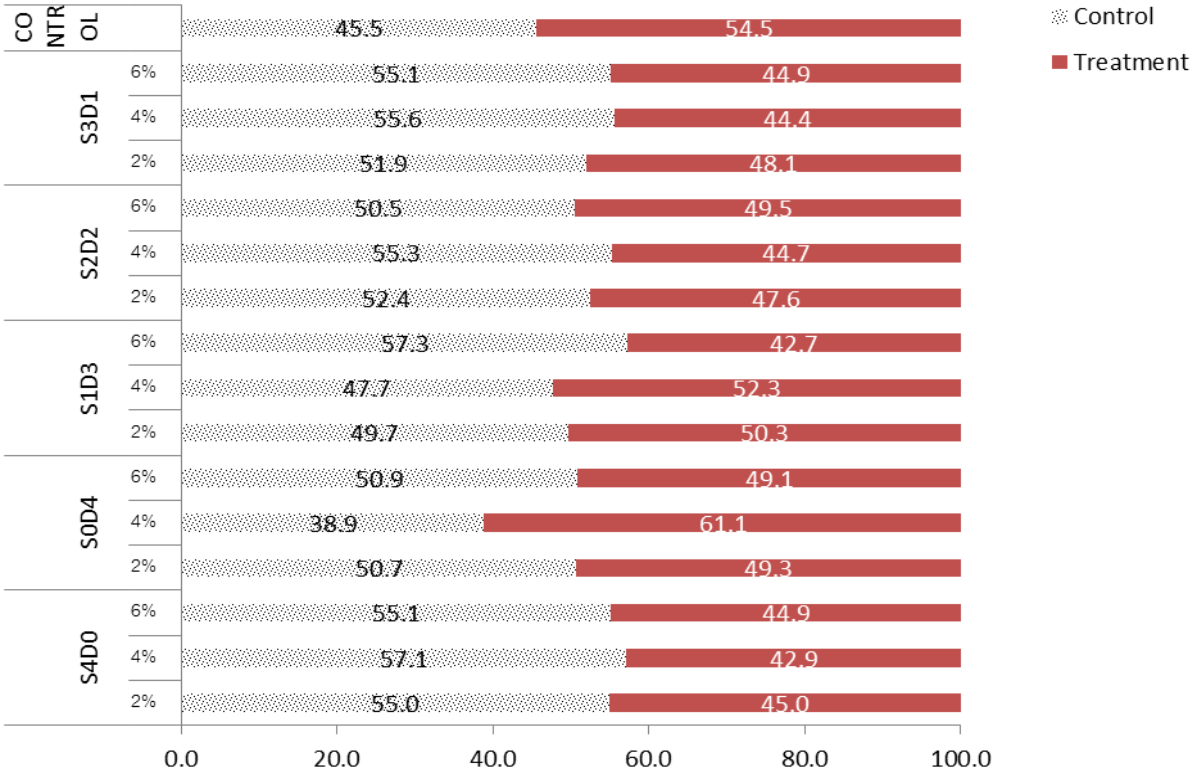

Figure 4 Number of adults of saw-toothed grain beetle, *Oryzaephilus surinamensis* L. developed from egg after treated with essential oil formulas.

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