Evaluation of essential oils against *Sitophilus zeamais* (Motshulsdy) (Coleoptera: Curculionidae)

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**ABSTRACT**

The objective of the current study was to determine the fumigant toxicity of essential oils for control stored product insects pest. Essential oils from ten plant species currently found in Thailand: pine (*Pinus Palustris*), lemon grass (*Cymbopogon citratus* Stapf), peppermint (*Mentha Piperita*), citronella grass (*Cymbopogon nardus* Linn), sweet acacia (*Acacia farnesiana*), cinnamon (*Cinnamomum verum* J.S. Presl), sweet orange (*Citrus sinensis* Pers), basil (*Ocimum basilicum* L.), clove (*Syzygium aromaticum* L.), and star anise (*Illicium verum* Hook) were extracted by steam distillation and tested for their insecticidal activities against maize weevil (*Sitophilus zeamais* Motshulsdy). Fumigant toxicity test was evaluated on adult of the maize weevil under laboratory conditions. Mortality of the maize weevil was observed and recorded every 12 h until 72 h. Responses varied with the test applied 100 and 10 µl of essential oils from the ten plants species on the tested insects. Three of the essential oils (sweet acacia, basil and star anise) showed high toxicity and were selected for the residuality test to mortality by contact with a surface of treated petri dish and glass jar. The results revealed that diluted 7.5 µl and 205.0 µl of essential oils from three plants (star anise, basil and sweet acacia) achieved a high mortality of the tested insects at 100% within 36 h after exposure to surface of treated petri dish and glass jar respectively.

**Keywords:** Essential oils, fumigant toxicity, maize weevil *Sitophilus zeamais* (Motshulsdy), mortality.

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**INTRODUCTION**

According to most of food products lost to various pests during post-harvest storage, consumers demand more natural processed products with long shelf-life but without chemical preservatives. Maize (*Zea mays* L.), rice and wheat are the three most important cereal crops worldwide (Regnault, 1997). Maize damage by *Sitophilus zeamais* causes food loss, increased poverty, and lower nutritional values of grain, increased malnutrition, reduced weight and market values (Keba and Sori, 2013). And also *S. zeamais* reduced germination percentage and maize production as most farmers in developing countries store grain and seed together (Pingali and Pandey, 2001).

Maize weevil, *Sitophilus zeamais* (Motschulsky), is a serious pest of economic importance in stored products in tropical and subtropical countries; infestation often starts in the field, but serious damage is done during maize storage (Suleiman et al., 2015; Fikremariam et al., 2009; Muzemu et al., 2013). It is the damage to grain by feeding activities of the adults and the development of immature stages within the grain. This not only reduces the grain quality but also produces a considerable amount of grain dust mixed with frass (Longstaff, 2010). It causes to weight loss of 20 to 90% for untreated maize in tropical countries (Giga et al., 1991). Maize is stored in commercial structures, with proper monitoring of
temperature and moisture content to control pests in
developed countries. But maize is often stored in
traditional structures with no environmental control and
usually without chemical protectants and usually without
chemical protectants in tropical countries (Dhilliwayo and
Pixley, 2003). Fumigant such as methyl bromide and
phosphine are still the most effective for the protection
from insect infestation of stored food, feedstuffs, and
other agricultural commodities (EPA, 2001). Some stored
product insects are found to have developed resistance
to methyl bromide and phosphine (Champ and Dyte,
1977).

The use of natural compounds from plants instead
synthetic chemical pesticides is an alternative that can
reduce the agriculture impact on the environment
(Vanichpakorn et al., 2010). The choice of native species
as source of oil and/ or extracts employed in pest control
could be a strategy to their sustainable use by local
communities, and consequently contribute to their
conservation. Plant may provide potential alternatives to
used for control insect agents because they constitute a
rich source of bioactive chemicals (Wink, 1993).

Numerous plants have been reported to have a variety of
biological activities against insects including insecticidal,
repellent, antifeedant, fumigant, growth regulatory,
antiovipostion activities (Isman, 2006; Ukeh et al., 2010).
Moreover, plant based insecticides often contain a
mixture of active substances, which can delay or prevent
resistance development (Wang et al., 2007). Plant
products can be used for insect pest control in form of
essential oils. Aim of this study was carried out to
evaluate the fumigant toxicity from ten plants species and
three essential oils were assessed against adults of
maize weevil, Sitophilus zeamais Motschulsdy under
laboratory conditions.

**MATERIALS AND METHODS**

**Insect preparation**

Maize weevil, Sitophilus zeamais (Motschulsky), was collected from
maize storage silos in Phitsanulok province, Thailand, and
reproduced in 1000 ml plastic containing maize (Zea mays L.) as
a source of food. The insects were maintained in the container at a
room temperature of 30 ± 1°C and 75 % RH. They were laboratory-
reared with laid eggs on maize, one week after laying eggs, the
insect parents were removed. The eggs were kept in the same
condition until adult emergence. Ten to fourteen-day olds of adults
were used for bioassay tests.

**Plants extract preparation**

The essential oils of ten plant species: pine (Pinus palustris), lemon
glass (Cymbopogon citratus Stapf), peppermint (Mentha piperita),
citronella grass (Cymbopogon nardus Linn), sweet acacia (Acacia
farnesiana), cinnamon (Cinnamomum verum J.S. Presl), sweet
orange (Citrus sinensis Pers), basil (Ocimum basilicum L.), clove
(Syzygium aromaticum L.), and star anise (Illicium verum Hook)
were extracted from aerial parts of the plants by steam distillation
using distilled water (Vogel et al., 1997). Subsequently, the oils
were collected in glass recipient and kept in amber colored glass
containers at 4°C until the subsequent assays. Pure essential oils
were employed in all the tests. The samples were subjected to
maize weevil S. zeamais (Motschulsdy). TWEEN 80 was used for
eulsion to stabilize the essential oils before testing.

**Insects bioassay test**

**Fumigant toxicity assay**

This bioassay employed the methodology of Pires et al. (2006)
cited by Jessica et al. (2010) which consisted of applying 0
(control), 1000, 100 and 10 μl of the ten essential oils on Whatman
N°10 filter paper (Whatman, Maidstone, Kent, UK), which were cut
into 3-cm diameter pieces and fixed under the petri dish. Filter
papers were impregnated with a series of concentrations of each
essential oil. The same procedure was used for the control with
filter paper without treatment and placed on the petri dish and then
the ten unsexed adult insects was placed on the petri dish after the
oils evaporated (10 insects/petri dish). There were four replicates of
each treatment. The experimental units were kept in laboratory at a
room temperature of 30 ± 1°C. Assessments of mortality were
made at 12, 24, 36, 48, 60 and 72 h of exposure.

**Mortality by contact with a surface of the treated container
(petri dish)**

The methodology of Kouninki et al.(2007) was used. The high
toxicities three from ten essential oils were selected to evaluate
doses of oils were star anise (Illicium verum Hook), basil (Ocimum
basilicum L.) and sweet acacia (Acacia farnesiana). The diluted of
the three essential oils, applying 0 (control), 2.5, 5.0 and 7.5 μl
(using the filter paper diffusion method with exposed into petri dish).
A solution of essential oils in acetone (99% purity), at the required
concentration of each treatment was applied on petri dish with 5 g
of maize grain. Each petri dish was infested 10 unsexed adult
insects, and stored in a room temperature of 30 ± 1°C. Each
treatment was carried out in 4 replicates. The mortality was
assessed at 12, 24, 48 and 72 h exposure to the toxic.

**Mortality by contact with a surface of the treated container
(glass jar)**

The methodology of Kouninki et al. (2007) was used, with slight
modifications that consisted of using 2600 cm² glass jar (using the
filter paper diffusion method with exposed into the jar) and applying
68, 137 and 205 μl instead of 2.5, 5.0 and 7.5 μl, respectively.
A solution of the three essential oils in acetone, at the required
concentration applied on 6 cm of filter paper and place on the jar.
The jar was agitated for 1 min for the oil to cover the interior
surface. The oil was allowed to evaporate at ambient temperature
for 1 h, then 10 unsexed adult insects were placed in each jar with
5 g of maize. Four replicates were made per treatment. The
substrates were kept at room temperature at 30 ± 1°C. Insect
mortality was assessed at 24, 48 and 72 h of exposure to the
essential oils.

As the mortality rate in the control was lower than 5%, this was
corrected with the Abbott formula (Abbott, 1925). An insect was
considered dead when there was no movement after prodding it
with a dissection needle.

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\frac{% \text{Mortality} = \frac{\% \text{test mortality} - \% \text{control mortality}}{100-\% \text{control mortality}} \times 100}
\]
Statistical analysis

The significance of treatments was calculated by one way Analysis of Variance (ANOVA) and effective treatment was separated by the Duncan’s New Multiple Ranges Test (DMRT). Differences between means were considered significant at P < 0.05.

RESULTS AND DISCUSSION

Fumigant toxicity of 10 essential oils against maize weevil Sitophilus zeamais (Motshulsdy)

Fumigant toxicity of ten essential oils with 100 and 10 µl as shown in Tables 1 and 2. The mortality of S. zeamais increased with the increase of volume of the oils and exposure time. Almost of the essential oils show fumigant toxicity with high mortalities ranging from 82.5 to 100% at 72 h after treatment with 100 µl (Table 1). When the decrease essential oils to 10 µl were applied on tested insects, the high fumigant toxicity occurred on the three essential oils against S. zeamais with significant differences (P < 0.01). Only three of essential oils showed significantly higher fumigant toxicity against S. zeamais than the other treatment with mortality of 100% at 72 h, whereas the other essential oils could not achieve up to 100 % insect mortality. Sweet acacia (Acacia farnesiana), basil (Ocimum basilicum L.) and star anise (Illicium verum Hook) achieved 100 % mortality of S. zeamais within 36 h after treatment. No mortality was observed in the untreated controls (Table 1). Therefore the three essential oils were selected to apply for testing mortality by contact with a surface of treated container with petri dish and glass jar as shown on Tables 3 and 4, respectively.

In general, mortality increased with increased exposure time to the essential oil, which concurs with Bittner et al. (2008). The 10 µl of essential oils from sweet acacia, basil and star anise exceeded 100% mortality at 36 h (Table 2). Particularly, the sweet acacia caused 100% mortality at 24 h after treatment, meantime mortality of basil and star anise was 87.5%. Similarly, it was observed in the fumigant action bioassay on S. zeamais (Motshulsly) by Jessica et al. (2010) that 35 µl of Peumus boldus Molina oil in a volume of 0.15 L has a rapid toxic effect, producing 100% mortality in 6 h. At 24 h, the treatments higher than 20 µl of the essential oil in 0.15 L caused 100% mortality. Also the mortality of S. zeamais after 24 h exposure to different dosage of C. dinisia oil demonstrated by Vedovatto et al. (2015), Vogel et al. (2005) reported that the essential oil of Rosmarinus officinalis L. and Eucalyptus blakelyi Maiden had fumigant action against the mite Tetranychus urticae Koch (Miresmailli et al., 2006); Sitophilus oryzae L. and Tribolium castaneum Herbst (Lee et al., 2003).

Table 1. Fumigant toxicities of 10 essential oils (100 µl) on mortality (%) Sitophilus zeamais Motshulsdy.

| Treatment              | % Mortality |
|------------------------|-------------|
|                        | 12 h        | 24 h        | 36 h        | 48 h        | 60 h        | 72 h        | df |
| Control (water)        | 0           | 0           | 0           | 0           | 0           | 0           | 0  |
| Pine oil               | 15.0        | 22.5        | 70.0        | 87.5        | 97.5        | 97.5        | *  |
| Lemon grass            | 0           | 0           | 7.5         | 7.5         | 10.0        | 12.5        | ns |
| Peppermint             | 92.5        | 100.0       | 100.0       | 100.0       | 100.0       | 100.0       | *  |
| Citronella grass       | 5.0         | 17.5        | 22.5        | 35.0        | 47.5        | 55.0        | ns |
| Sweet acacia           | 55.0        | 75.0        | 95.0        | 100.0       | 100.0       | 100.0       | *  |
| Cinnamon               | 22.5        | 45.0        | 62.0        | 69.5        | 79.5        | 84.5        | *  |
| Sweet orange           | 65.0        | 70.0        | 77.5        | 80.0        | 80.0        | 82.5        | *  |
| Basil oil              | 80.0        | 97.5        | 97.5        | 97.5        | 97.5        | 97.5        | *  |
| Clove Oil              | 32.5        | 57.5        | 82.5        | 82.5        | 82.5        | 82.5        | *  |
| Star anise             | 70.0        | 85.0        | 97.5        | 100.0       | 100.0       | 100.0       | *  |

ns = non significant; * = significant difference, means (followed by the same letter) are not significantly different at 5% level by DMRT.

Mortality by contact with a surface of treated container (petri dish) with the 3 selected essential oils

The three selected of essential oils (sweet acacia, basil and star anise) were applied on petri dish, the mortality by contact with a surface of the treated petri dish showed mortality increases with increases volume of the essential oil. At 12 h, S. zeamais response to the sweet acacia oil with mortality was 75, 92.5 and 95% at treatment of 2.5, 5.0 and 7.5 µl respectively. However, star anise oil given the low mortality to S. zeamais only 22.5 and 30.0% at treatment of 2.5 and 5.0 µl respectively, but sharply increase reached a mortality to 100% at treatment of 7.5 µl at the first 12 h after testing. There was no mortality in the untreated control (Table 3). In 2003, some researcher
Table 2. Fumigant toxicities of essential oils (10 µl) on *Sitophilus zeamais* Motshulsdy.

| Treatment               | % Mortality | df  |
|-------------------------|-------------|-----|
|                         | 12 h        | 24 h | 36 h | 48 h | 60 h | 72 h |
| Control (water)         | 0           | 0    | 0    | 0    | 0    | 0    | ns  |
| Pine oil                | 2.5         | 15.0 | 17.5 | 17.5 | 17.5 | 17.5 | ns  |
| Lemon grass             | 2.5         | 2.5  | 5.0  | 7.5  | 7.5  | 10.0 | ns  |
| Peppermint              | 30.0        | 32.5 | 37.5 | 40.0 | 50.0 | 70.0 | *   |
| Citronella grass        | 2.5         | 5.0  | 5.0  | 7.5  | 10.0 | 15.0 | ns  |
| Sweet Acacia            | 95.0        | 100.0| 100.0| 100.0| 100.0| 100.0| n.s.|
| Cinnamon                | 17.5        | 22.5 | 32.5 | 40.0 | 52.5 | 52.5 | *   |
| Sweet Orange            | 0           | 0    | 0    | 0    | 7.5  | 17.5 | ns  |
| Basil oil               | 62.5        | 87.5 | 100.0| 100.0| 100.0| 100.0| *   |
| Clove Oil               | 10.0        | 15.0 | 32.5 | 35.0 | 47.5 | 50.0 | ns  |
| Star anise              | 50.0        | 87.5 | 100.0| 100.0| 100.0| 100.0| *   |

ns = non significant; * = significant difference, means (followed by the same letter) are not significantly different at 5% level by DMRT.

Table 3. Mortality (%) of *Sitophilus zeamais* Motshulsdy exposed to petri dish surface treated with the 3 selected essential oils.

| Treatment               | % Mortality | df  |
|-------------------------|-------------|-----|
|                         | 12 h        | 24 h | 36 h | 48 h | 60 h | 72 h |
| Control (water)         | 0           | 0    | 0    | 0    | 0    | 0    | ns  |
| Sweet acacia oil 2.5 µl | 75.0        | 82.5 | 82.5 | 85.0 | 87.5 | 87.5 | *   |
| Basil oil 2.5 µl        | 12.5        | 30.0 | 60.0 | 62.5 | 75.0 | 75.0 | *   |
| Star anise oil 2.5 µl   | 22.5        | 32.5 | 52.5 | 55.0 | 60.0 | 62.5 | *   |
| Sweet acacia oil 5.0 µl | 92.5        | 97.5 | 100.0| 100.0| 100.0| 100.0| *   |
| Basil oil 5.0 µl        | 20.0        | 44.5 | 69.5 | 74.5 | 74.5 | 79.5 | *   |
| Star anise oil 5.0 µl   | 30.0        | 67.5 | 97.5 | 100.0| 100.0| 100.0| *   |
| Sweet acacia oil 7.5 µl | 95.0        | 100.0| 100.0| 100.0| 100.0| 100.0| *   |
| Basil oil 7.5 µl        | 35.0        | 55.0 | 100.0| 100.0| 100.0| 100.0| *   |
| Star anise oil 7.5 µl   | 100.0       | 100.0| 100.0| 100.0| 100.0| 100.0| *   |

* = significant difference, means (followed by the same letter) are not significantly different at 5% level by DMRT.

Table 4. Mortality (%) of *Sitophilus zeamais* Motshulsdy exposed to 2600 cm³ glass jar surface treated with the 3 selected essential oils.

| Treatment               | % Mortality | df  |
|-------------------------|-------------|-----|
|                         | 12 h        | 24 h | 36 h | 48 h | 60 h | 72 h |
| Control (water)         | 0           | 0    | 0    | 0    | 0    | 0    | ns  |
| Sweet acacia oil 68.0 µl| 27.5        | 72.5 | 82.5 | 82.5 | 82.5 | 82.5 | *   |
| Basil oil 68.0 µl       | 67.5        | 85.0 | 97.5 | 100.0| 100.0| 100.0| *   |
| Star anise oil 68.0 µl  | 72.5        | 75.0 | 90.0 | 97.5 | 100.0| 100.0| *   |
| Sweet acacia oil 137.0 µl| 47.5       | 62.5 | 92.5 | 97.5 | 97.5 | 97.5 | *   |
| Basil oil 137.0 µl      | 57.5        | 82.5 | 100.0| 100.0| 100.0| 100.0| *   |
| Star anise oil 137.0 µl | 57.5        | 77.5 | 100.0| 100.0| 100.0| 100.0| *   |
| Sweet acacia oil 205.0 µl| 90.0        | 95.0 | 95.0 | 97.5 | 97.5 | 97.5 | *   |
| Basil oil 205.0 µl      | 87.5        | 90.0 | 100.0| 100.0| 100.0| 100.0| *   |
| Star anise oil 205.0 µl | 100.0       | 100.0| 100.0| 100.0| 100.0| 100.0| *   |

ns = non significant; * = significant difference, means (followed by the same letter) are not significantly different at 5% level by DMRT.
Figure 1. The effect of the 3 selected essential oils on % mortality of *Sitophilus zeamais* Motshulsdy exposed to petri dish surface (a) and exposed to 2600 cm³ glass jar surface (b).

Figure 2. Percentage mortality of *Sitophilus zeamais* Motshulsdy exposed to various periods of time to the 3 selected essential oils on filter paper discs.
Mortality by contact with a surface of treated container (glass jar) with the 3 selected essential oils

Additional, the treated container was slightly modified that consisted of using 2600 cm³ glass jar instead of the petri dish and applying of three selected essential oils was 68.0, 137.0 and 205.0 µl. Essential oil of star anise at treatment of 205.0 µl resulted the highest of mortality of 100% at 12 h. No mortality was observed in the untreated controls (Table 4). As the time increases between observations the mortality of adults S. zeamais, mortality of all treatments reached at 100% at 36 h. Therefore, the mortality rate that was obtained by contact with a surface of treated container of petri dish or glass jar with the 3 selected essential oils given highest at 36 h. Aslan et al. (2004) reported that the level of mortality has been reached at 48 h with the essential oils of Achillea biebersteinii Afan and A. wilhelmsii, and at 96 h with oil of Pistacia spp. (Aslan et al., 2004).

These experimental were conducted to determine whether the insecticidal activity of the 3 selected essential oils were attributable to fumigant activity. In all cases, considerable differences in insect mortality were noted with different doses and exposure times as shown on Figures 1 and 2. It can be concluded that for control of S. zeamais, higher doses for a relatively short period are much more effective than lower doses for longer periods. Fumigant toxicity of the three essential oils to bruchid increase of exposure time from 6 to 48 h resulted in an increase of larval mortality, whilst further increases of exposure time gave no additional detrimental effect (Papachristos and Stamopoulos, 2002).
Suleiman R, Rosentrater KA, Bern CJ, 2015. Evaluation of maize weevils *Sitophilus zeamais* Motschulsky. Infestation on seven varieties of maize. J Stored Prod Res, 64: 97-102.

Ukeh DA, Birkett MA, Bruce TJA, Allan EJ, Pickett JA, Mordue Luntz AJ, 2010. Behavioral responses of the maize weevil, *Sitophilus zeamais*, to host (maize grain) and non-host plant volatiles. Pest Manag Sci, 66: 44-50.

Vanichpakorn P, Vanichpakorn WD, Cen XX, 2010. Insecticidal activity of five Chinese medicinal plants against *Plutella xylostella* L. larvae. J Asia-Pacific Entomol, 13: 169-173.

Vedovatto F, Valério Júnior C, Astolfi V, Mieliczki PAA, Roman SS, Paroul N, Cansian RL, 2015. Essential oil of *Cinnamodendron dinisii* Schwанke for the control of *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). Rev Bras Plantas Med, 17(4) supl.3 Botucatu.

Vogel H, Razmilic I, Doll YU, 1997. Contenido de aceite esencial y alcaloides en diferentes poblaciones deboldo (*Peumus boldus* Mol.). Ciencia e Investigación Agraria, 24:1-6.

Vogel H, Razmilic I, San Martin J, Doll U, González YB, 2005. Plantas medicinales chilenas., 192 p. Editorial Universidad de Talca, Talca, Chile.

Wang YN, Shi GL, Zhao LL, Liu SQ, Yu TQ, Clarke SR, Sun JH, 2007. Acaricidal activity of Juglans regia leaf extracts on *Tetranychus viennensis* and *Tetranychus cinnabarinus* (Acar: Tetranychidae). J Econ Entomol, 100: 1298-303.

Wink M, 1993. Production and application of phytochemicals from an agricultural perspective. In: van Beek, T.A., Breteler, M. (Eds.), Phytochemistry and Agriculture, Vol.34. Clarendon, Oxford, UK, pp. 171-213.

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