

Insecticidal effect of three essential oils and Beauvericin nano gel on *Sitophilus oryzae* and *Sitophilus granarius* (Coleoptera :Curculionidae)

Magda M. A. Sabbour

ABSTRACT

Sitophilus oryzae and *Sitophilus granarius* (Coleoptera :Curculionidae) harmful insects cause a damages to stored product seeds. These pests controlled by chemical materials which pollute the environments. Essential oils are a natural compound used as insecticide to a lot of pests. In our research we controlled these pests by essential oils, *Cinnamomum zeylancium*, *Zingiber officinale*, *Cuminum cyminum* and Beauvericin nano gel 2%. Results showed that, the LC_{50s} of *S. Oryzae* were 38, 44, 151 and 18 ppm for Beauvericin nano gel 2% *C. zeylancium*, *C. cyminum* and *Z. officinale*, respectively. Also LC_{50s} for *S. granarius* obtained by the corresponding treatments were, 41, 144, 11 and 29 ppm. The accumulative of *S. oryzae* significantly increased after 48 (H) by 2.8 fold after application of *Cinnamomum zeylancium* oil. When *Zingiber officinale* Oil applications the accumulative mortality increased by 3,3fold. The Beauvericin nano gel 2% showed that the accumulative increased by increasing the exposure time. *Cinnamomum zeylancium* showed a higher effect on the wheat weevil *S. granarius*, where the accumulative mortality recorded 91 after 48 (H). The lower accumulative mortality after Beauvericin nano gel 2%, recorded 38 after 48(H). The repellency test showed that, the highest repellent found among the Beauvericin nano gel 2% treatments. Data presented show that the persistence effectiveness of oil significantly decreased by increasing the storage time. The potential of the insecticidal oils was high after 45 days of storage, against *S. oryzae* and *S. granarius*. The potential of the insecticidal effect of oil recorded the lowest percentage at the end of storage after 100 days to both *S. oryzae* and *S. granarius*.

Key words: *Cinnamomum zeylancium*, *Zingiber officinale*, *Cuminum cyminum* nano gel, persistence, control

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INTRODUCTION

The pest infestation of the stored grains is one of the main problems which causing damage of about ten % of the food crops stored grains in the world. The rice weevil *Sitophilus oryzae* and *Sitophilus granarius* (Coleoptera: Curculionidae) are among the common types that attack stored grains. They are serious grain insects because the female live more about 2 years and lay eggs reach to 290 to 300 eggs (Hong *et al.*, 2018). They attack several economic stored seeds in the field and store by strong moth parts and chewing the seed

causing holes. These pests cause a seeds loss and decrease its quality (Boudreaux, 1969). These pests developed inside the grains seed then the adults emerged leave crushed and destroyed seeds behind Trematerra *et al.* (1999). Some chemical pesticides were used to control these insects of the stored. These chemical materialsharmful to humans due to the toxic effect of them. Natural Pesticides that are safe for humans and the environment. In recent years, many researchers have been interested in searching for alternatives safe materials to control these insects (Mostafa,

1999; Ismail *et al.*, 2014, 2015; Sabbour and Abdel-Raheem, 2016, Sabbour *et al.*, 2020; Sabbour and Shadia, 2020) Including the use of natural products of plants as a successful and effective natural control of their qualities. Desirable as they are rapidly degradable and highly effective compounds against harmful insects and little harm to (Peterson *et al.*, 2000). Use plant parts extract act as buffers to feed insects, repellents, or growth regulators (Mustafa, 1999, Abdel-Raheem *et al.*, 2015, 2019). Beauvericin extracted from fungi, has insecticidal, repellent and antibiotic effects on insects (Logrieco *et al.*, 2002; Mohamed Abdel-Raheem *et al.*, 2020; Sabbour, 2019). As a result of the above, this study aimed to assess the effect of nano gel Beauvericin and by using three powdered essential oils in two *Sitophilus* beetles species insect by knowing its fatal and repellent effect the insect as alternatives to chemical pesticides, reduce pollution and protect the environment.

MATERIALS AND METHODS

Insect rearing

Cereal weevils, *Sitophilus oryzae*, and *S. granarius* adults were collected and preserved at a temperature of 25 ° C, in the Insect Laboratory, Department of Pests and Plant Protection. A week for the purpose of adapting it to laboratory conditions before conducting the experiment. Then make artificial diet and reared the two pests on it according to Sabbour (2012). The artificial diet consists of wheat powder, glycine (16%) yeast powder (7%) and one drops of antibiotic. The last all compounds mixed together then feed the newly larvae in the laboratory of NRC.

Extraction of essential oil

Cuminum cyminum, *Cinnamomum zeylancium* and *Zingiber officinale* seeds extracted their essential oils by hydro distilled. The tested seeds (100 g) taken then grinded, put in a flask (one L), then covered with boiled water around (100° C). After six hours the volatile oil were collected in the reservoir Sabbour and Shadia Abd El-Aziz (2016a), the same obtained by Sabbour and Shadia Abd El-Aziz (2016). Also these experiments

protocol followed by Sabbour and Shadia Abd El-Aziz (2020). After the extraction of oils, remove the excess of water by Sabbour and Nayera (2019a,b) (Sahaf *et al.*, 2007).

The tested essential oils tested at the concentrations of 20, 40, 60, 100 and 120 ppm. Small pieces of foams dipped in each of the last concentrations, then left for drying. The foam put between (500gm) of rice seeds for *S. oryzae*, and wheat seeds for *S. granarius* in glass container (tree L) covered with muslin, left till the end of the experiment Sabbour and Shadia Abd El-Aziz (2016a, 2016b).. The control experiments left without any treatments. The number of the dead beetles in each jar was counted every day and the percentages of mortality were corrected by using the Abbott formula (Abbott, 1925), the LC_{50s} were determined through the probit analysis (Finney 1971). The experiment was carried under laboratory conditions; 25± 3°C and 65 ± 5 R.H %. All of experiment were replicated five times.

Sublethal on the accumulative mortality

The effect of the sublethal dose of the Beauvericin 2% at 11 ppm. The beetles of both *S. oryzae* and *S. granarius* treated by sublethal dose of three tested oils at 12 for the oil *Cinnamomum zeylancium* and 42 ppm for the oil, *Zingiber officinale* and *Cuminum cyminum* at 22 ppm. Twenty five beetles of both tested *S. oryzae* and *S. granarius*, were exposed to sprayed disk of foam at the last concentrations mentioned for 7, 17, 24, 36 and 48 hrs. The mortality estimated and determined till the end of the experiments.

Preparation of nano gels

Beauvericin Nano gels were prepared by self-assembly prepared method by of Chen *et al.* (2003). The nanoparticles preparations made by Fusaric acid –modified beauvericin. Dissolving 1000mg Fusaric acid, 50 mL dimethyl sulfoxide, (DMSO, Sigma Aldrich, C99.9 %, Mw = 78.13 g/mol). Beauvericin (2 g) obtained from (Sigma Aldrich company) then added 2 % acetic acid solution (200 mL).

Persistence bioassay

LCs75 which obtained from the last made experiments used to persistence calculations of the oil and nano gel Beauvericin 2%. Thirty of adult beetles from the two *Sitophilus* species were introduced to each vial every three days of the experiments. The mortality counted every day after exposure. The persistence experiment was continued until 100 days and the essential oils and Beauvericin nano gel 2% lost their insecticidal effects.

Repellency test

The repellency experiments were made by choice test used an arena chambers. A tested oils were applied at the concentrations of (1%) sprayed on a filter paper (Whatman No. 1) the nit placed in the cell A. A filter paper sprayed by distilled water put in the next arena B for control experiments. Thirty of the newly hatched beetles were put in each of the last designed arenas. Then , the number of beetles found in each arena cells A and B were detrained each 1, 3,5, 7 days. The repellency percentages numbers calculated by the following Equation: $D = [1 - (T/C)] \times 100$ (Lwande *et al.*, 1985) where T and C represent the mean number of beetles in cells A and B treated and untreated, respectively.

RESULTS

Data found in table 1, show that the LC₅₀ of the tested two oils and nano gel Beauvericin on *Sitophilus oryzae*. Results proved that LC₅₀ of Table 2. Accumulative mortality of *Sitophilus oryzae* and *Sitophilus granarius* exposed to the tested essential oils and Nano gel at 5%.

Beauvericin nano gel 2% decreased by 1.1 fold that LC₅₀ of *Cinnamomum zeylancium* and decreased by 3.9 and 8.3 of *Cuminum cyminum* compared by *Zingiber officinale* LC₅₀ (Table 1).

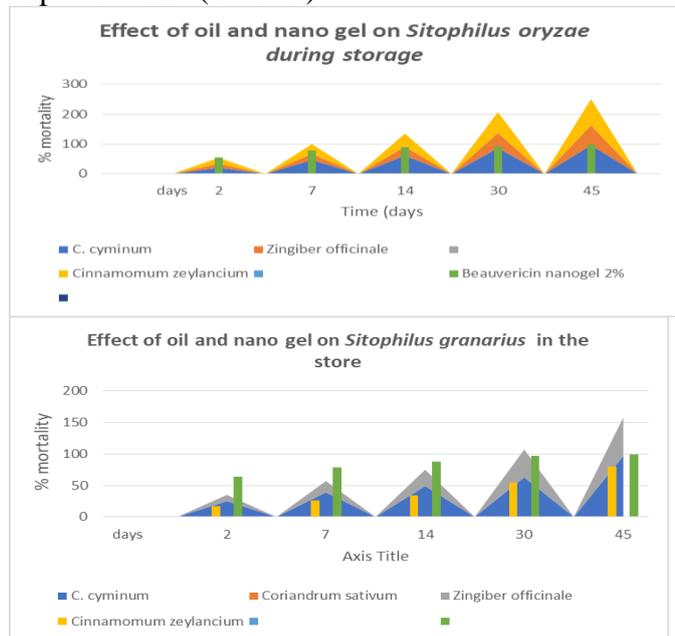
Table 1. Effect of the tested oils and nano-gel chitosan 5% on *Sitophilus oryzae* and *Sitophilus granarius*

Tested substance	LC ₅₀ (ppm)	Confidence limits 95%
<i>Sitophilus oryzae</i>		
<i>C. zeylancium</i>	44	12-78
<i>Z.officinale</i>	151	141-177
Beauvericin nano gel 2%	38	11-54
<i>C. cyminum</i>	28	20-48
<i>Sitophilus granarius</i>		
<i>C. zeylancium</i>	41	22-68
<i>Z. officinale</i>	144	140-167
Beauvericin nano gel 2%	29	10-64
<i>C. cyminum</i>	20	10-44

The effect of the tested essential oils and nano gel on *S. granarius*, showed that the LC₅₀ of Beauvericin nano gel 2% decreased by 1.4 fold of LC₅₀ of *Cinnamomum zeylancium*, by 12.8 fold of *Cuminum cyminum* and 4.9 fold of LC₅₀ of *Zingiber officinale* (Table 1). The accumulative mortality of *S. oryzae* showed in (Table 2).

Insect	Tested materials time (H)	Concentrations (ppm) at Sublethal dose				
		7	14	24	36	48
<i>Sitophilus oryzae</i>	<i>C. zeylancium</i>	28	35	48	59	80
	<i>Z. officinale</i>	19	22	33	42	63
	Beauvericin nano gel 2%	7	11	23	35	37
	<i>C. cyminum</i>	20	43	62	82	93
	F value/Lsd5%	34/19.8				
<i>Sitophilus granarius</i>	<i>C. zeylancium</i>	23	38	50	78	91
	<i>Z. officinale</i>	17	26	44	68	81
	Beauvericin nano gel 2%	8	11	23	37	38
	<i>C. cyminum</i>	22	45	66	84	96
	F value/Lsd5%	22/16.4				

The accumulative of *S. oryzae* significantly increased after 48 (H) by 2.8 fold after application of *C.zeylancium* oil. When *Zingiber officinale* Oil applications the accumulative mortality increased by 3.3 fold. The Beauvericin nano gel 2% showed that the accumulative increased by increasing the exposure time (Table 2).



C. zeylancium showed a higher effect on the wheat weevil *S. granarius*, where the accumulative mortality recorded 91% after 48 hrs the lower accumulative mortality after Beauvericin nano gel 2% , recorded 38 after 48 hrs (Table 2). The repellency test showed in figures 1 and 2, the highest repellent found among the Beauvericin nano gel 2% treatments. Figure 2. Repellency of the oils and nano gel on *S. granarius* in storage period the lowest repellent on *S. oryzae* and *S. granarius* was the oil *Z. officinale*.

The persistence of essential oils and nano gel Beauvericin is presented in Fig. 3, 4. The data presented show that the effectiveness of oil significantly decreased by increasing the storage time. The potential of the insecticidal oils was high after 45 days of storage, against *S. oryzae* and *S. granarius*. The potential of the insecticidal effect of oil recorded the lowest percentage at the end of storage after 100 days to both *S. oryzae* and *S. granarius* (Fig. 3 and 4).

Fig.3 .persistence of tested oils in the store on *S. oryzae*.

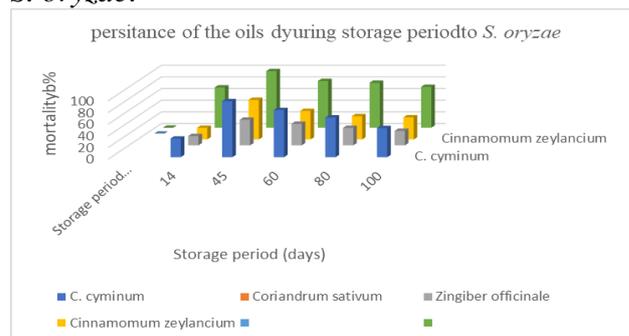
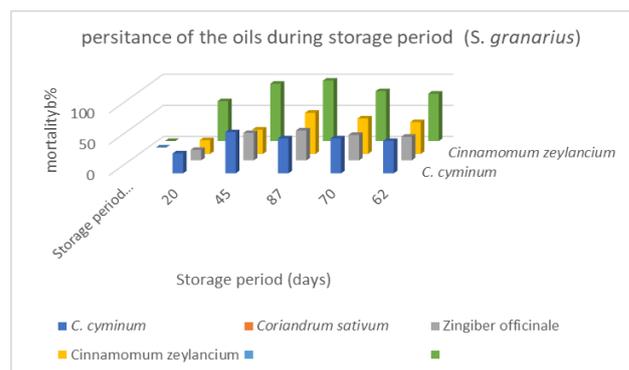


Fig.4 .persistence of tested oils in the store on *S. granarius*.



DISCUSSIONS

Results show that the decrease in LC₅₀of the tested Beauvericin to 38 and 29 PPM for *S. oryzae* and *S. granarius*, respectively. These results agree with Sabbour (2007), how found that the LC₅₀ of bioagent used decreased this agree with Sabbour and Shadia (2015). Our results showed highly confirmed with Sabbour (2013a and b) who reported that the essential plant oils have a strongly repellency effect an *S. oryzae* and *Ephestia Kühniella* in both laboratory and store. Su (1989) who strongly confirmed that, when using acetone extract for *Myristica fragrans* which showed a strongly repellency on rice weevil, *S. oryzae* L. using a nine essential oils extract *C. zeylancium* which protect the wheat seeds from yellow corn weevil *S. zeamais* infestations. Nat *et al.* (2013) mentioned that, the aqueous extracts are an exothermic efficacy and strong repellency on stored product insects. Ishii *et al.* (2010) stated that, when using five plant extract including *Cinnamomum*, *Cinnamon* *Zingiber officinal*

and oils and ginger excelled at *S. zeamais*. They reported that *C. zeylanicum* have their effective repellency over the rest of the plants, followed by *Z. officinale* (Sabbour *et al.*, 2012; Sabbour and Shadia, 2010; Sabbour, 2020a,b,c). (Odili and Epedi, 2008) study of the efficacy of *C. zeylanicum* on *Tribolium castaneum* and found that it has a strongly repellent effect against the target weevils. A studying the effect of four powders of plants including ginger vs. They mentioned *C. zeylanicum* good in controlling *S. oryzae*. They reported that all of the plant extracts used are useful for preserving rice from *S. oryzae* L (Sabbour, 2020 a,b,c). The variation in the mortality ratios between the plants of this study is due to a difference in the chemical components. For different plants or for a difference in the way the same plant is used as a powder or aqueous extract (hot or cold) or alcoholic extract. Mixing the grains with vegetable powders causes the insects to die due to this. Until the powder minutes stick to insect bodies absorbing water from them or the insect friction with them causes. It results in the removal of the waxy layer from the wall of its bodies (Sabbour and Shadia, 2017) where water evaporates, dries and dies (Al-Azzawi and Muhammad, 1983), and the reason for the difference in the halos is due to the variation of the active substance present in the plant powders (Sabbour *et al.*, 2012; Sabbour and Nayer, 2020). Shaaban and Al-Mallah (1993) reported that the variation in mortality rates may be due to toxic effect through contact with it The powder to the surface of the body and the penetration of the chemical compounds of kyotalk by penetrating into the elastic areas in it or through respiratory openings causing paralysis and rapid death, as it is believed that the cause of the effect is caused by containment Plants on secondary alkaline, phenolic, and terpene sub-clocoside substrates and potent compounds that work. As a preventive or repellent impediment lead to the inhibition of the process of laying eggs and the proportion of hatching and in the process of role slaughter, while Saadi (2001) explained that 64 lost larvae and adult deaths

when treated *Callosobruchus maculatus* (Fab.) % of southern cowpea beetle adults. In concentration, 4% g eucalyptus powder is due to its effect on the insect's nervous and digestive systems (Halawa *et al.*, 1998). While there are highly significant differences in the characteristics of killing and expulsion between concentrations of plant powders Used in the study. The results of this study were consistent with the findings of Khalaf and Ilan (2002). Al-Farhan and Khalaf (2009) reported that the increased mortality of adult insect beetle due to increased plant concentrations. Its exerting effect when increasing concentrations of powders mixed with grains. All constituents of the tested essential oils *Cinnamomum zeylanicum*, *Cuminum cyminum*, *Zingiber officinale*, belong to were terpenes. Which have an insecticidal effect on insects causing a toxicity by fumigant, contact, and ingestion action on stored product insect pests (Prates *et al.*, 1998; Lee *et al.*, 2003; Rozman *et al.*, 2007; Abdelgaleil *et al.*, 2009 and Sabbour 2020 c). The rapid penetration of the tested essential oils, to in the insect body, cause death to them. The essential oils found in *C. cyminum* cause inhibitions to enzymes activity to *S. oryzae* (Abdelgaleil *et al.*, 2009).

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MAGDA M. A. SABBOUR

Pests and Plant Protection Dept. National Research Centre, 33rdElBehouse St. El-Tahrir Street, Dokki, P.O. Box 12622, Giza, Egypt.
 Fax: 002 33370931; **E-mail** : sabbourm@yahoo.com