Sabbour and Abd El-Aziz *Bulletin of the National Research Centre* (2019) 43:80
https://doi.org/10.1186/s42269-019-0129-3

Bulletin of the National Research Centre

RESEARCH Open Access

Impact of certain nano oils against *Ephestia kuehniella* and *Ephestia cutella* (Lepidoptera-Pyralidae) under laboratory and store conditions

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Abstract

Background: For the purpose of environmental safety, many essential oils were recommended in many Integrated Pest Management (IPM) programs to protect plants from insect's infestations and have few effects on non-target organisms and the environment. This study was aimed to investigate and estimate the activity of four nano essential oils against tested insects under laboratory and store conditions.

Methods: The nano encapsulation process was carried out by polymerization technology. The tested nano oils were experimented at tested concentration (0.5%) for their insecticidal activities against the third-instar larvae of tested insects. After 7 days of exposure, accumulative mortality percentages were calculated in the treated and untreated control. Also, the tested nano oils were sprayed to the foam granules and were mixed with wheat (2 g foam/100 g wheat) for testing the oviposition inhibitory effects of tested oils. Additionally, the experiment was designed to test the persistent effect of tested oils against *Ephestia kuehniella* and *E. cautella* on foam as surface protectant after 90-day intervals.

Results: The larval accumulative mortality of tested insect species enhanced progressively with the increase of exposition times (7 days). In the case of tested insects, rosemary oil gave the highest accumulative mortality percentages. The persistence effects of tested nano oils on foam and covering gunny bags showed various biological activates by reducing number of eggs, almost suppressing the percentages of progeny emergence (F1) and high percentages of malformation of tested insects. The efficacy of the tested nano oils on the weight loss of wheat grains after 90 days of storage was experimented.

Conclusion: Results of this work and that of other researchers indicate that some essential oils might be useful for managing insects in enclosed spaces because of their fumigant action. Plant essential oils and their active ingredient have potentially high bioactivity against a range of insects. Furthermore, they are highly selective to insects. Incorporation of essential oils into a controlled release nanoformulation prevents rapid vaporization and degradation, increases constancy, and preserves the lower effective dosage/application. Treated foam with nano rosemary and nano eucalyptus oil and covering gunny bags provided many efficient effects against tested insects.

Keywords: Camphor, Rosemary, Anise, Garlic, Nano-oil

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Background

The synthetic pesticides and their residual problems have increased the necessity for efficacious biodegradable pesticides with greater selectivity. Substitution strategies have inclusive the search for new sorts of pesticides. Several studies have focused on the potential use of botanicals applications in biological control of different insect pests since some are selective, biodegrade to nontoxic products, and have few effects on non-target organisms and the environment (Singh and Upadhyay 1993; Abd El-Aziz and Sharaby 1997; Isman 2000; Abd El-Aziz and Ezz El-Din 2007; Kim et al. 2010). The action of essential oils against stored-product insects has been extensively studied (Negahban et al. 2006; Sahaf and Moharramipour 2008; Sabbour and Abd-El-Aziz 2010; Abd-El-Aziz 2011; Sabbour and Abd-El-Aziz 2017; Sabbour and Abd-El-Aziz 2018). In spite of the fact that essential oils have the most promising properties, problems related to their volatility, poor water solubility, and potential for oxidation have to be resolved before being used as alternative pest control means (Moretti et al. 2002).

Each of nano pesticides, nano fungicides, and nano herbicides have been utilized in agriculture (Owolade et al. 2008; Sabbour 2013; Sabbour and Abd-El-Aziz 2016a, 2016b; Sabbour and Solieman 2016; Sabbour and Abd-El-Aziz 2017). Incorporation of essential oils into a controlled release nano formulation prevents rapid vaporization and degradation, increases constancy, and preserves the lower effective dosage/application (Ghormade et al. 2011).

Therefore, the present study aimed to estimate the activity of four nano essential oils against tested insects under laboratory and store conditions.

Methods

Tested insect

Larvae of *Ephestia kuehniella* and *Ephestia cautella* (third instar) were used in the experiments. The target insects were reared under laboratory conditions on semi-artificial diet (fine wheat with some adherent endosperm) with 20% glycine and 5% yeast powder.

Tested nano essential oils

Four essential oils were used in the bioassay, rosemary (*Rosemarinus afficinalis*), eucalyptus (*Eucalyptus globulus*), garlic (*Allium sativum*), and anise (*Pimpinella anisum*). The essential oils were obtained by steam distillation methods of dried plants (Guenther 1961). The tested oils were prepared according to (Sabbour and Abd-El-Aziz 2016a). The nano encapsulation process was carried out by polymerization technology. The tested oils were used as a core material, and urea (U) and formaldehyde (F) as shell materials. Sulfuric acid solution (10% *w*/*w*) was prepared in our laboratory to control the pH (4.4) of emulsion, and tween 80 (polysorbate 80) was used as emulsifier (Merck, Germany). The obtained suspension of nano capsules was cooled down to ambient temperature, rinsed with deionized water, filtered, and finally dehydrated by freeze-drying using a LIO-5P, which is a Freeze Dryers for Laboratory Use. (Apparatus CinquePascal, Trezzano SN, Milan, Italy).

The insecticidal activities of tested Nano- oils

The tested nano oils were experimented at tested concentration (0.5%) for their insecticidal activities against the third-instar larvae of tested insects. According to Abd El-Aziz (2001), the foam granules were sprayed with the tested nano oils, left to dry, and mixed with wheat (2 g foam/100 g wheat). Four glass jars as replicates were used. Thereafter, ten third-instar larvae were introduced into each glass jar which was covered with muslin for suitable ventilation. In the case of untreated control, 12 replicates were kept under the same conditions without any treatments. After 7 days of exposure, accumulative mortality percentages were calculated in the treated and untreated control. All tests were carried at 27 ± 2 °C and 65 ± 5% relative humidity (RH). The number of dead larvae in each jar was assessed, and the percentages of mortality were calculated. The experiment was repeated four times.

The ovipositional deterrent effects of the tested nano oils

The tested nano oils were sprayed to the foam granules and were mixed with wheat (2 g foam/100 g wheat) for testing the oviposition inhibitory effects of tested oils (Abd El-Aziz 2001). Two pairs of mixed sex of tested moth adults (2–3 days old) were placed with treated or untreated wheat grains with foam particles in glass jars (250 cc capacity) covered with muslin. The moths were left to lay eggs, and then the numbers of deposited eggs on treated or untreated wheat/female were calculated in the tested jars. Percentages of adult emergence (F1) and adults' malformations were calculated. Four glass jars as replicates were used, and the test was repeated three times (Abd El-Aziz et al. 2000).

The persistence effect of tested oils in the store

Experiment was designed to test the persistent effect of tested oils against insect species on foam as surface protectant after 90-day interval. All gunny sacks (20 × 20 cm each) were full of heat-sterilized wheat (100 g each), fastened, each with a string. The foam granules (about 0.5 cm in diameter) were sprayed with treatments, dried, and provided as a layer between sacks. Two of newly emerged moths of each tested insect species were placed in a jar (2 l capacity with four gunny sacks). After 90 days, the percentage of grain weight lost was calculated from the differences between the original and the final weight in each jar using the following formula:

$$\%\mathrm{WL}\ (\%\mathrm{Weight\ Loss}) = [(\mathrm{OW} - \mathrm{FW})/\mathrm{OW}] \times 100$$

where OW is the original weight and FW is the final weight. Each experiment was repeated five times (Abd El-Aziz and Abd El-Ghany 2018).

Statistical analysis

The data was analyzed using analysis of variance (ANOVA, one way), where significant differences between the treatments were observed. Mean values were significantly separated by using the least significant difference (LSD) test at 5% level (Sokal and Rohlf 1981).

Results

The insecticidal activities of tested nano oils

The accumulations mortality of tested insects larvae during the first week (7 days) were subjected in (Table 1). The larval accumulative mortality of tested insect species enhanced progressively with the increase of exposition times (7 days). In the case of *E. cautella* and *E. kuehniella*, rosemary oil gave the highest accumulative mortality percentages (86.7% and 81.3%) after 7 days from treatment followed by eucalyptus treatment which amounted to 69.9 and 65.3%, respectively, and the lowest percentage (48.9% and 43.3%) was recorded in the case of anise oil. Garlic oil recorded moderate accumulative mortality (40.48% and 50.8%) in the case of *E. cautella* and *E. kuehniella*, respectively. There were highly significant differences between untreated control and all the treatments. After 7 days, there was no significant difference between rosemary oil and eucalyptus oil, while there were highly significant differences between rosemary oil and garlic and anise oils.

Table 1 Accumulative percentage mortality of tested insect larvae (third instar) during 1 week in treated foam with different nano oils

Tested oil	Time (days)	*E. kuehniella*	*E. cautella*
Nano rosemary	0	45.30	46.60
	2	53.90	58.30
	4	64.30	67.60
	7	81.30	86.70
Nano garlic	0	29.50	29.26
	2	35.37	36.90
	4	48.50	39.50
	7	50.80	40.48
Nano eucalyptus	0	31.10	33.20
	2	45.30	46.90
	4	58.50	59.30
	7	65.30	69.90
Nano anise	0	27.30	26.20
	2	30.30	30.20
	4	39.30	38.90
	7	43.30	48.90
Untreated	0	0.00	0.00
	2	0.00	0.00
	4	0.00	0.00
	7	0.20	0.10
F test		18.5	22.7
Lsd5%		8.7	9.9

The ovipositional deterrent effects of the tested nano oils

The persistence effects of tested nano oils on foam and covering gunny bags showed various modes of action by reducing number of eggs, which almost suppressed the percentages of progeny emergence (F1) and high percentages of malformation of tested insects (Tables 2 and 3). The oviposition was extensively inhibited. The highly reduction in the number of eggs was recorded in the case of nano rosemary (96.53% and 96.54) in *E. kuehniella* and *E. cautella*, respectively. There were highly significant differences between untreated control and all the treatments in the case of the tested insect species. There were no significant differences between treatments. The highest number of laid eggs was recorded in the case of nano anise oil (31.4 ± 1.71 and 44.4 ± 1.21 eggs) in the case of *E. kuehniella* and *E. cautella*, respectively, compared with the untreated control (299.4 ± 1.88 and 297.9 ± 7.89 eggs). There was a high reduction in % adult emergence (F1) in the case of all treatments; the recorded percentage of adult malformation was 100% in the case of nano rosemary and nano eucalyptus in *E. kuehniella*, while in *E. cautella* 98% in rosemary and eucalyptus, 99% in garlic, and 78% in anise was recorded.

The persistence effect of tested oils in the store

The efficacy of the tested nano oils on the weight loss of wheat grains after 90 days of storage is clarified in Figs. 1 and 2). All tested nano oils were significantly ($P < 0.05$) better than the untreated control. The least weight loss was recorded in the case of nano rosemary and nano eucalyptus (2%), followed by nano garlic (6%) and nano anise (10%) compared with the untreated control (38%) in the case of *E. kukeinella*. While in the case of *E. cutella*, least weight loss was recorded in the case of nano rosemary (7%) and followed by nano eucalyptus (8%), nano garlic (11%), and nano anise (17%) compared with the untreated control (43%).

Discussion

The main constituent of many plant extracts and essential oils are mainly monoterpenoids (Ahn et al. 1998).

Table 2 Effect of nano oils on number of laid eggs/female, % adult malformation, and % of adult emergence (F1) of *E. kuehniella* during storage

Tested materials	No. of eggs/♀ ± S.E.	% adult emergence (F1)	% adult malformation
Nano rosemary	10.4 ± 9.89 (96.53)	1.0	100.0
Nano eucalyptus	21.4 ± 7.81 (92.85)	9.0	100.0
Nano garlic	19.4 ± 1.87 (93.52)	6.0	99.0
Nano anise	31.4 ± 1.71 (89.51)	12.0	90.0
Control	299.4 ± 1.88	100.0	0.0
F test	38.7		
Lsd5%	19.9		

*numbers between brackets represent percent reduction than control

Monoterpenoids are typically volatile and rather lipophilic compounds that can penetrate into insects rapidly and interfere with their physiological functions (Lee et al. 2002). Essential oils are efficient against storage pests because oils have a slippy and/or oily property in which the eggs of the insect could not be attached to the grain surface. It can also have a repellent action whereby the insect cannot come in contact with the grain. The minimum grain damage in oil treatments might be due to the reduction in the number of adult emergences which results in less weight loss percentage and less kernel damage (Haili 2006). Moreover, Yang et al. (2009) tested the activity of nanoparticles and free garlic essential oil against red flour beetle. They mentioned that the control effectiveness of the nanoparticles of garlic oil was 80% followed by free garlic oil (11%). The efficient components of nanoparticles turned out to be highly efficient due to their delayed and constant release. So, integral essential oils into a controlled release nano formulation stop fast vaporization and degradation, enhance persistence, and maintain the lower effective dosage/application (Ghormade et al. 2011).

The larval accumulative mortality percentages of tested insect species enhanced progressively with the increase of exposition times (7 days). In the case of *E. cautella* and *E. kuehniella*, rosemary oil gave the highest accumulative mortality percentages (86.7% and 81.3%) after 7 days from treatment followed by eucalyptus which amounted to 69.9 and 65.3%, respectively, and the lowest percentage (48.9% and.43.3%) was recorded in the case of anise oil. The main constituents of many essential oils are monoterpenoids. They have fumigant activity because of their high volatility which might be of importance for controlling insects of stored-products (Ahn et al. 1998). These results are in agreement with those of Konstantopoulou et al. (1992), Regnault-Roger and Hamrouni (1995), and Ahn et al. (1998). Purslane oil (bulk and nano) caused a highly significant increase in the mean mortality percent followed by castor oil against larvae of *E. kuehniella* (Sabbour and Abd-El-Aziz 2016b). The mortality percentage of *Callosobruchus analis* beetles reached 97.5% after being treated with castor oil (Aheer et al. 1996). Nanoemulsion was more efficient than *Mentha longifolia* essential oil against *E. kuehniella* Zeller (Louni et al. 2018). Moreover, nanoemulsion has powerful contact toxicity and influence on mortality rate of *E. kuehniella* fifth-instar larva. Also, Shahmirzaei et al. (2016) mentioned that fumigant toxicity of *M. longifolia* oil is powerful than its contact toxicity. Using the technique of nanoemulsion can increase the contact activity of essential oil and its strength, out of slow-release of active ingredients over time.

The persistence effects of tested nano oils on foam and covering gunny bags showed various biological activates by reducing number of eggs, almost suppressing the percentages of progeny emergence (F1)

Table 3 Effect of nano oils on number of laid eggs/female, % adult malformation, and % of adult emergence (F1) of *E. cutella*during storage

Tested materials	No. of eggs/♀ ± S.E.	% adult emergence (F1)	% adult malformation
Nano rosemary	10.3 ± 0.0 (96.54)	0.1	98.0
Nano garlic	20.4 ± 1.21 (93.15)	2.0	99.0
Nano eucalyptus	20.4 ± 5.38 (93.15)	1.0	98.0
Nano anise	44.4 ± 1.21 (85.51)	11.0	78.0
Control	297.9 ± 7.89	100.0	0.0
F test	36.7		
Lsd5%	18.7		

*numbers between brackets represent percent reduction than control

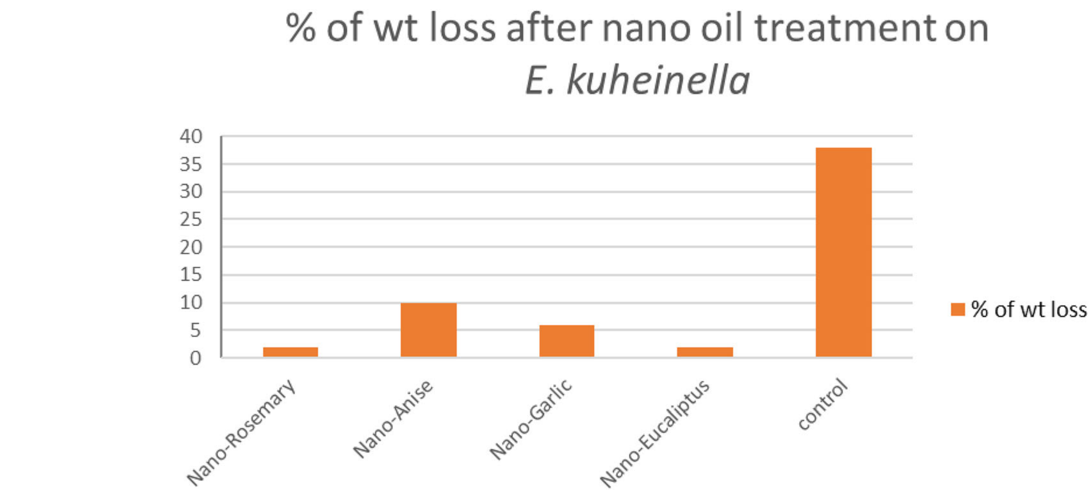

Fig. 1 Percentage of grain weight loss after nano oil treatment on *E. kukeiella*

and highly percentages of malformation of tested insects. A complete inhibition of adult emergence of *E. cautella* was subjected in case of nano purslane oil. This can be referred to the sterilizing effect of purslane oil (bulk and nano) on the adults as well as its toxicity to the laid eggs and adult emergence during storage intervals up to 125 days (Sabbour and Abd-El-Aziz 2016b). Spraying cotton plants with 2.5% of white mustard oil caused reduction in egg laying. The adult moths laid only 7% of their egg masses, and the repellency % was 89.4% (Abd El-Aziz and Sharaby 1997). Nano purslane oil completely suppressed the % of emerged moths (0.00%) compared with the untreated control (66%) (Sabbour and Abd-El-Aziz 2017). The tested essential oils have a repellent action whereby the insect cannot come in contact with the grain.

All tested nano oils have highly significant differences ($P < 0.05$) than the untreated control. The least weight loss was recorded in the case of nano rosemary and nano eucalyptus (2%), followed by nano garlic (6%) and nano anise (10%) compared with the untreated control (38%) in the case of *E. kukeinella*. While in the case of *E. cutella*, least weight loss was recorded in the case of nano rosemary (7%) and followed by nano eucalyptus (8%), nano garlic (11%), and nano anise (17%) compared with the untreated control (43%). In laboratory experiments, grain losses by insects of stored products had been tested (Moino et al. 1998; Padina et al. 2002; Sabbour and Abd El-Aziz 2007; Arya and Tiwari 2013). Up to 60 days, the percentages of infestation did not exceed, 13, 15, 20, and 20%, respectively. Mostly, the weight loss for treatments was increased with the increase of the storage intervals. The highest percentage of damaged

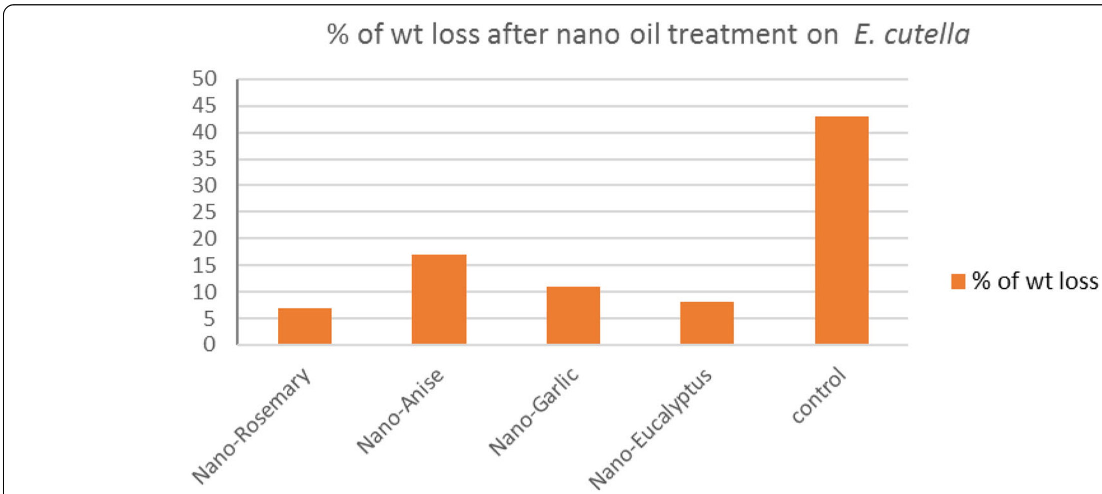

Fig. 2 Percentage of grain weight loss after nano oil treatment on *E. cutella*

seeds was recorded in the untreated plots. After 5 months, *Beauveria bassiana*, *Metarhizium anisopliae*, mustard, and nigella treatments had a significantly lower percentages of grain damage and lower seed weight loss (10, 15, 17, and 22%), respectively (Sabbour and Abd El-Aziz 2007). These results are in agreement with those of Arya and Tiwari (2013). Nothing is more effective than neem, jatropha seed, mustard, and cow dung ash which had no weight loss of seeds, while cow dung powder recorded the least weight loss (4.80%) in comparison to untreated control (34.80%). Abd El-Aziz and Abd El-Ghany (2018) studied the impact of Diatomaceous Earth Modifications to control granary weevil. They mentioned that the least weight loss was listed for Al-DE treatment (7.0%) followed by Ca-DE (10.0%), Na-DE (13.0%), and the original-DE (25.0%) compared with the control (38%).

Conclusion

The conclusion of the results of this work and that of other researchers indicate that some essential oils might be useful for managing insects in enclosed spaces because of their fumigant action. Furthermore, they are highly selective to insects. Incorporation of essential oils into a controlled release nano formulation prevents rapid vaporization and degradation, increases constancy, and preserves the lower effective dosage/application. Treated foam with nano rosemary and nano eucalyptus oils and covering gunny bags provided many efficient effects against *E. cautella* and *E. kuehniella*. The present experiments obviously showed that tested nano oils have been found significantly efficient against tested insects as more insect mortality, a smaller number of eggs, less adult emergence, high percentage of adult malformation, and less percentage of weight loss after 90 days of storage. These results established the criteria on which to base future experiments under simulated farmer storage practice.

Abbreviations

Al-DE: Aluminum diatomaceous earth; Ca-DE: Calcium diatomaceous earth; DE: Natural diatomaceous earth; *E. cautella*: *Ephestia cutella*; *E. kuehniella*: *Ephestia kuehniella*; Na-DE: Sodium diatomaceous earth

Acknowledgements

The authors are very grateful to Pests and Plant Protection Department and National Research Centre for all the facilities during this work.

Funding

Not applicable.

Availability of data and materials

All dataset on which abstracted of the study have been drawn are presented in the main manuscript. All tables or figures have not been published anywhere else before. All data and materials are available.

Authors' contributions

SMM contributed in putting the idea, designed the experiments, made the laboratory and store experiments, took part in the statistical analysis and writing the research, and reviewed the manuscript. ASE, also, contributed in putting the idea, designed the experiments, made the laboratory and store experiments, took part in the statistical analysis and writing the research, and reviewed the manuscript. Both authors read and approved the final manuscript.

Ethics approval and consent to participate

The manuscript does not contain studies involving human participants, human data, or human tissue. Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Publisher's Note

Received: 12 March 2019 Accepted: 10 May 2019
Published online: 22 May 2019

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