



Acaricidal and repellent activities of essential oil of *Eucalyptus globulus* against *Dermanyssus gallinae* (Acari: Mesostigmata)

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ABSTRACT

Introduction: By considering an increase in drug resistance against red mites, finding the nonchemical herbal acaricide against *Dermanyssus gallinae* (De Geer) (Acari: Mesostigmata) is necessary to kill them and to reduce the chemical resistance against chemical acaricides in this specie. *Dermanyssus gallinae* is a potential vector of the causal agent of several viral diseases such as Equine encephalitis and St. Louis encephalitis. It can be a vector of bacteria such as *Salmonella* spp., *Mycobacterium* spp. and *Erysipelothrix rhusiopathiae*. It is also known to cause itching dermatosis in humans. In this study acaricidal and repellent activities of essential oil of *Eucalyptus globulus* against *Dermanyssus gallinae* were studied.

Methods: After extracting the essential oil, different concentrations of the plant extract were prepared. Then, acaricidal effect of different concentrations was tested on poultry red mite, *Dermanyssus gallinae*, by dropping 3-4 drops of essential oil on mites. Repellent activity of essential oil was tested by Y-tube olfactometer bioassay. After the test, total number of killed and repellent mites reported.

Results: Concentration of 1:2 or 50% had more acaricidal effect on mites. Also essential oil of *Eucalyptus globulus* had repellent activity against red mites.

Conclusion: This study showed that essential oil of *Eucalyptus globulus* had acaricidal and repellent activities against red mites. Hence it might be used as a herbal acaricide against it to kill and to reduce the chemical resistance in this specie.

Implication for health policy/practice/research/medical education:

The *Eucalyptus globulus* essential oil possesses acaricidal and repellent activities against red mites and might be used instead of chemical substances against this specie.

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Introduction

It has been estimated that about 2.5 million tons of pesticides are used on crops each year and the worldwide damage caused by pesticides reaches \$100 billion annually. The reasons for this are: (a) the high toxicity and non-biodegradable properties of pesticides and (b) the residues in soil, water resources and crops that affect public health (1). Thus, on the one hand, one needs to search the new highly selective and biodegradable pesticides to solve the problem of long term toxicity to mammals and, on the other hand, one must study the environmental friendly

pesticides and develop techniques that can be used to reduce pesticide use while maintaining crop yields. Natural products are an excellent alternative to synthetic pesticides as a mean to reduce negative impacts to human health and the environment (1). The move toward green chemistry processes and the continuing need for developing new crop protection tools with novel modes of action make discovery and commercialization of natural products as green pesticides an attractive and profitable pursuit that is commanding attention (2). The concept of "Green Pesticides" refers to all types of nature-oriented and ben-

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eficial pest control materials that can contribute to reduce the pest population. They are safe and ecofriendly. They are more compatible with the environmental components than synthetic pesticides (1). Thus in the present concept of green pesticides, some rational attempts have been made to include substances such as plant extracts, hormones, pheromones and toxins from organic origin and also encompass many aspects of pest control such as microbial, entomophagous nematodes, plant derived pesticides, secondary metabolites from microorganisms, pheromones and genes used to transform crops to express resistance to pests (2). Essential oils are defined as any volatile oil(s) that have strong aromatic components and that give distinctive odor, flavor or scent to a plant. These are the by-products of plant metabolism and are commonly referred to as volatile plant secondary metabolites. Essential oils are found in glandular hairs or secretory cavities of plant-cell wall and are present as droplets of fluid in the leaves, stems, bark, flowers, roots and/or fruits in different plants (2). The aromatic characteristics of essential oils provide various functions for the plants including (a) attracting or repelling insects, (b) protecting themselves from heat or cold; and (c) utilizing chemical constituents in the oil as defence materials, as fragrances and flavorings in the perfume and food industries, respectively, and more recently for aromatherapy and as herbal medicines (3).

The genus *Eucalyptus* includes many species which are widely distributed in different regions of Iran. The eucalyptus oil is a complex mixture of a variety of monoterpenes and sesquiterpenes, aromatic phenols, oxides, ethers, alcohols, esters, aldehydes and ketones (4). Essential oil of *Eucalyptus* has been placed under GRAS (Generally regarded as safe) category by Food and Drug Administration (FDA) of USA and classified as nontoxic (5). Component 1, 8-cineole is the main constituent of eucalyptus oil (6) that insecticidal properties have been studied on large storage pests (7). Fumigation toxicity of essential oil and extracts of various species of *Eucalyptus* on storage pest has been studied by several researchers (8).

The poultry red mite, *Dermanyssus gallinae* (De Geer) (Acari: Mesostigmata), is a parasite with wide host-range, including wild and domestic birds. It is known from 30 bird and 10 mammal species (9). In the absence of birds, *D. gallinae* will also attack mammals such as rodents, dogs, cats, horses, as well as humans (10). *Dermanyssus gallinae* is a potential vector of the causal agents of several viral diseases such as Equine encephalitis (11) and St. Louis encephalitis (12). It can be a vector of bacteria such as *Salmonella* spp. (13), *Mycobacterium* spp. and *Erysipelothrix rhusiopathiae* (14). It is also known to cause itching dermatosis in humans (15). The most frequently used acaricides against *D. gallinae* are organophosphates, carbamates and pyrethroids. Organophosphates and carbamates are toxic to arthropods and mammals by virtue of their ability to inactivate the enzyme acetylcholinesterase (16). There are reports of *D. gallinae* resistance to permethrin and organophosphates (17). In order to prevent the development of resistance, groups of acaricides are sug-

gested to be used in rotation (18).

So finding the nonchemical herbal acaricides against *Dermanyssus gallinae* (De Geer) (Acari: Mesostigmata) is necessary to kill them and to reduce the chemical resistance against chemical acaricides in this specie. In this study acaricidal and repellent activities of essential oil of *Eucalyptus globulus* against *Dermanyssus gallinae* were studied.

Materials and Methods

Preparation of essential oil

The plant materials, ie, leaves were collected from the areas around Isfahan province called Najafabad in center of Iran in June and July 2014. The plant was identified and authenticated as *Eucalyptus globulus* in the Herbarium of the Research Institute of Jahade Keshavarzi of Shahrekord, Iran and a herbarium specimen was deposited there. Then leaves were dried in an oven equipped with warm air circulation. Two hundred grams of the air-dried materials were ground and powdered. The powder was subjected to hydro-distillation for 3 hours using a Clevenger-type system. Finally essential oil extracted. The oil was kept at 4°C in a brown vial.

Collection of the tested mites

Mostly hand searching technique, infested birds and simple nets were used for collecting mites. Hand searching refers to simply getting down on the birds skin and on the surface of birds nest. Most of the mites were obtained from under feathers of head, chest, vent and other parts of bird's bodies. Most of the old bird nests are suitable for this technique. In addition to this, some of the mites were collected from fissures of nest's walls and cages. Finally about 500 healthy mites were collected and transferred immediately to laboratory. Before the test, about 15 mites were died that removed from the tests.

Evaluating the acaricidal activity of essential oil

Different concentrations of the essential oil were first prepared. To do this, 5 ml of the plant extract with 5 ml of water were used for 1:2 or 50% proportion, 5 ml of plant extract with 15 ml of water for 1:4 or 25% proportion, 5 ml of the plant extract with 20 ml of water for 1:5 or 20% proportion, 5 ml of the plant extract with 45 ml of water for 1:10 or 10% proportion, 5 ml of the plant extract with 95 ml of water for 1:20 or 5% proportion, and 5 ml of the plant extract with 495 ml of water for 1:100 or 1% proportion. In addition a negative control group was used by water without any essential oil and positive control group treated with 5 g organophosphates (chemical industrial powder) diluted with 5 ml water. All of the above groups tested in triplicate by 20 mites.

Evaluation of the repellent activity of essential oil

Repellent activity of essential oil of *Eucalyptus globulus* against *Dermanyssus gallinae* was studied by Y-tube olfactometer bioassay. Y-tube olfactometer consists of a glass Y-tube with a main arm (the stem) and 2 arms containing one repellent and a control in other one, where a low rate

air movement is created by sucking the air in the two arms of the Y-tube with a pump connected to the stem. The essential oil sample and control are applied on a paper attached to the arms of the tube. Insects are introduced into the tube by a hole located at the center (the joint point of the three tubes). After introduction, the hole is closed with a rubber stopper and the pump is operated. After 2 minutes of exposition, the number of insects on each of the 2 tubes (treated and control) are scored to assess the percentage of repellency (3). In this study, 50 mites were put into every arms and 10 ml essential oil was added into treated tube and nothing into control tube. Two minutes after pumping, the number of repellent mites was reported as repellent percentage of treated and control groups.

Data analysis

The data were analyzed by one-way analysis of variance (ANOVA) and significant differences between means were assessed by Tukey test using SPSS version 19.0. $P < .05$ was considered as significantly difference.

Results

Acaricidal activity of different concentrations of *Eucalyptus globulus* essential oil is shown in Table 1. In essential oil group, 93.8% mites (*Dermanyssus gallinae*) repelled and in negative control group 2.33% mites repelled in Y-tube olfactometer ($P < .05$). Concentration of 1:2 or 50% had more acaricidal effect on mites. There was no significant difference between concentration 1:4 or 25% and 1:5 or 20%. Concentration 1:20 or 5% and concentration 1:100 or 1% had less acaricidal effect, negative control had no killed mite and all of the mites in positive control were killed (Table 1).

Discussion

Essential oils from plants may be an alternative source of mosquito larval control agents, since they constitute a rich source of bioactive compounds that are biodegradable into nontoxic products and potentially suitable for use in integrated management programs. In fact, many researchers have reported the effectiveness of plant essential oils against mosquito larvae (19).

Results of this study showed that essential oil of *Eucalyptus globulus* had repellent activity against *Dermanyssus gallinae*. In this study concentration of 1:2 or 50% had more acaricidal effect on mites. There was no significant difference between concentration of 1:4 or 25% and 1:5 or 20%. Concentration 1:20 or 5% and concentration of

1:100 or 1% had less acaricidal effect.

Eucalyptus is one of the world's most important and most widely planted genera. It includes more than 700 species and belongs to the family of Myrtaceae (2). Many species of the genus Eucalyptus from the Myrtaceae family are used in China folk medicine for a variety of medical conditions. For examples, hot water extracts of dried leaves of *Eucalyptus citriodora* are traditionally used as analgesic, anti-inflammatory and antipyretic remedies for the symptoms of respiratory infections, such as cold, flu, and sinus congestion (20). Its main uses are the production of essential oils, which are used for medicinal and pharmaceutical purposes. In addition, *Eucalyptus camaldulensis* and *Eucalyptus urophylla* are also known to contain bioactive products that display antibacterial, antifungal, analgesic and anti-inflammatory effects, as well as antioxidative and antiradical activities (21,22).

Same as results of this study, repellent properties of essential oils and extracts from genus Eucalyptus are also well documented. These studies presented high repellency against *Ixodes ricinus*, *Aedes albopictus*, *Mansonia* and *P. humanus capitis*; low activity to *A. aegypti* and larvae of *C. pomonella*, and no effect on *L. serricornis* (cigarette beetle) (23). Repellents based on eucalyptus oils have been formulated and shown against *Leptoconops* biting midges (24). Also the pesticidal activity of eucalyptus oils has been due to the components such as 1, 8-cineole, citronellal, citronellol, citronellyl acetate, p-cymene, eucamalol, limonene, linalool, α -pinene, g-terpinene, α -terpineol, alloocimene, and aromadendrene (21).

Among the various components of eucalyptus oil, 1, 8-cineole is the most important one. In fact, it is a characteristic compound of the genus Eucalyptus which is largely responsible for a variety of its pesticidal properties and insecticide effects. 1, 8-cineol or Eucalyptol is a natural product found in the essential oil extracted from leaves of several species of Eucalyptus spp. (Myrtaceae), and it has already been confirmed for *Tribolium castaneum* (25).

Conclusion

Result of this study showed that essential oil of *Eucalyptus globulus* had strong effect against *Dermanyssus gallinae*. The current method for repellent arthropods which are based on chemotherapy, are limited and are not ideal because they are often associated with severe side effects. So it is possible to use the essential oil instead of chemical drugs. Results obtained from this study showing the acaricidal and repellent activities of essential oil of *Eucalyptus*

Table 1. Acaricidal activity of different concentrations of essential oil of *Eucalyptus globulus* against mites (*Dermanyssus gallinae*)

Groups	Mean number of killed mites \pm SEM						Negative Control	Positive Control
	1:2 or 50% of essential oil	1:4 or 25% of essential oil	1:5 or 20% of essential oil	1:10 or 10% of essential oil	1:20 or 5% of essential oil	1:100 or 1% of essential oil		
No. of killed mites \pm SEM ^a	18.87 \pm 3.12 a	16.25 \pm 2.71 b	15.14 \pm 1.74 c	9.62 \pm 1.41 d	2.53 \pm 2.68 e	1.16 \pm 0.82 e	0 \pm 0.00 f	20 \pm 0.00 g

^a Different letters within each row (ie, within each time interval) indicate significant differences ($P < .05$) between mean of killed mites number in each group according to Tukey test.

globulus against *Dermanyssus gallinae* may indicate the possible use of this essential oil as herbal acaricide.

Authors' contributions

ADS carried out the design and contributed to data analysis and writing and finalizing the manuscript and participated in the experiments. SMG and AZDS and KPK participated in most of the experiments and in manuscript preparation.

Conflict of interests

The authors declared no competing interests.

Ethical considerations

Ethical issues (including plagiarism, misconduct, data fabrication, falsification, double publication or submission, redundancy) have been completely observed by the authors.

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