BF-1940
[1-9]

RESEARCH ARTICLE Indian Journal of Animal Research

Evaluation of the Molluscicidal Effects of *Thymus vulgaris* and *Syzygium aromaticum* Oils *via* Contact and Bait Methods on *Monacha cartusiana* Snails with Special Emphasize on Neurotoxical, Biochemical, Histopathological and Endocrine Disrupting

Heba Abdel Tawab[1], Shawky M. Aboelhadid[2], Ebtesam A. Yousef[3]
Abdel-Azeem S. Abdel-Baki[1], Ahmed O. Hassan[4], Saleh Al-Quraishy[5], Heba Y. Ahmed[6]

10.18805/IJAR.BF-1940

ABSTRACT

Background: *Monacha cartusiana* is considered one of the important pests in agriculture and public health sectors. So, its management is critical to global food security. The purpose of this study was to look at the toxicity of *Thymus vulgaris* (thyme) and *Syzygium aromaticum* (clove) oil by bait and contact technique.

Methods: Gas Chromatography-Mass Spectrometry (GC-MS) was used to determine the essential oil's components. the LC_{50} of oils via bait and contact testing were determined in the laboratory experiment. Moreover, the molluscicidal activity following the exposure of snails to LC_{50} were evaluated through using snail soft tissues.

Result: The GC–MS analysis depicted that Thymol (33.89%) and eugenol (32.82%) was the major constituent present in the oils. both oils have ovicidal activity causing complete inhibition of eggs hatchability. Compared to control, the oils showed perturbations in antioxidant/oxidant biomarker, where reduced glutathione significantly decrease and lipid peroxidation increased. Concerning energy reserves biomarker, total lipid level significantly elevated in oils treated snails, While the total protein significantly decreased, Moreover, metabolic enzyme alkaline phosphatase was markedly augmentation. Both oils at the tested doses caused a significant inhibition in acetyl choline esterase activity and downregulation in 17β-estradiol and testosterone hormones. Also, snail digestive glands and ovotestis showed pathological alterations after essential oil exposure. In field application, thyme, clove and binary mixture caused significant reduction in *M. cartusiana* population by bait assay. Our findings emphasis thyme and clove oil's potential as an effective biorational molluscicide against pestiferous snails as an alternative to chemocentric management.

Key words: Biochemical, Biorational molluscicide, Clove, *Monacha cartusiana,* Neurotoxical, Thyme.

INTRODUCTION

Land snails are among the most dangerous pests attacking agricultural crops globally and their economic impact has grown significantly (Barker, 2002). *Monacha cartusiana*, one of these land snails, is regarded as a snail pest that seriously harms numerous agricultural crops. It has critical impacts in field crops, fruits, vegetables and ornamental plants nurseries (Abd El-Halim *et al*., 2021; Murshed *et al.,* 2024). In addition, it acts as a vector for a number of parasites and diseases that affect cultivated plants (Godan, 1983). Pest management in agriculture is crucial for global food security since it ensures crop productivity and quality production (Muhie, 2022).

Molluscicides are used extensively to control snail infestations all over the world. However, the indiscriminate application of synthetic molluscicides such as methomyl and other carbamates to control snails poses a serious risk to beneficial and non-target organisms and the emergence of pesticide resistance (Simms *et al*., 2002; Abdel-Halim *et al*., 2021; Pathak *et al*., 2022). To address all of the above issues, there is an urgent need to develop innovative, low-cost, eco-friendly and effective alternative molluscicides.

[1]Department of Zoology, Faculty of Science, Beni-Suef University, Beni Suef, Egypt.
[2]Department of Parasitology, Faculty of Veterinary Medicine, Beni-Suef University, Beni-Suef 62511, Egypt.
[3]Department of Zoology, Faculty of Science, Sohag University, 82524 Sohag, Egypt.
[4]Department of Medicine, Washington University School of Medicine, St. Louis, MO 63110, USA.
[5]Department Zoology, College of Science, King Saud University, Riyadh, Saudi Arabia.
[6]Department of Harmful Animals Research, Plant Protectin Research Insititute, Agriculture Research Center.

Corresponding Author: Heba Abdel Tawab, Department of Zoology, Faculty of Science, Beni-Suef University, Beni Suef, Egypt.
Email: hoba_abdo_2010@yahoo.com

How to cite this article: Tawab, H.A., Aboelhadid, S.M., Yousef, E.A., Abdel-Baki, A.A.S., Hassan, A.O., Al-Quraishy, S. and Ahmed, H.Y. (2025). Evaluation of the Molluscicidal Effects of *Thymus vulgaris* and *Syzygium aromaticum* Oils *via* Contact and Bait Methods on *Monacha cartusiana* Snails with Special Emphasize on Neurotoxical, Biochemical, Histopathological and Endocrine Disrupting. *Indian Journal of Animal Research*. 1-9. doi: 10.18805/IJAR.BF-1940.

Submitted: 16-12-2024 **Accepted:** 07-09-2025 **Online:** 08-10-2025

Plant-derived essential oils (EOs)and components are key sources of novel bioactive compounds with broad-spectrum insecticidal activity and they are typically regarded safe for humans and the environment (Dassanayake *et al*., 2021; Radwan and Gad 2021; Sekar *et al.,* 2025). EOs have been shown to be effective against a variety of pests, including insects, mites, fungi, slugs, snails and nematodes (Isman, 2000; Rana *et al.,* 2011; Yazdani *et al*., 2014; Sujatha *et al.,* 2015; Pavela and Benelli, 2016; Barua *et al*., 2017; Da Silva *et al.,* 2018; Klein *et al*., 2020; Nasiou and Giannakou, 2020).

Thymus vulgaris, "thyme," and *Syzygium aromaticum* (clove) are well-known aromatic herbs that have long been used as a spice and herbal remedy (Aljabeili *et al*., 2018; Haro-Gonzalez *et al*., 2021). The most prevalent fragrant bioactive compounds in thyme and clove essential oils are thymol and eugenol (Bozin *et al*., 2006; Gavaric *et al*., 2015; Pandey *et al.,* 2022). The US FDA claims that clove and thyme essential oils are both safe and have a favorable effect on the environment (Food *et al*., 2007).

The pilot study of Ismail and Abdel-Kader (2011) concluded that the flower-bud powder and commercially available eugenol of *Syzygium aromaticum* seem to have a possible role in the control of land snails. Also, Ali (2017) found that the aqueous extract of *Thymus vulgaris* have mollus-cicidal effect against the land snail *Eobanica vermiculate*.

This study expands on the earlier conclusion to examine the molluscicidal effects of thyme and clove against the land snail, *M. cartusiana*.

MATERIALS AND METHODS

Tested compounds and its consistent

Thyme and clove oils were purchased from Purity Factory in the food sector of Kom Abu-Radi Industrial Area, Alwasta, Beni-Suef, Egypt. methyl N- (methylcarbamoyloxy) ethanimidothioate, also known as Methomyl (20% SL), is a carbamate compound purchased from the chemical company of Egypt chem international Agricultural chemical, Egypt, Cairo (purity: 95.5%). The Ministry of Agriculture and Land Reclamation in Egypt has recommended using methomyl to fight land snail infestation on Egyptian fields.

Gas Chromatography-Mass Spectrometry (GC-MS) analysis was used to determine the essential oil's components. Chemical analysis of thyme and clove oils were carried out in the Central Laboratory of the Faculty of Postgraduate Studies for Advanced Sciences, Beni-Suef University, Egypt, using an Agilent Technologies GC system 7890A/5975 Inert MS with Triple Axis Detector. The constituent derivatives were identified by matching their mass spectra to derivative spectra in the Library Search Report [C:\Database\NIST11.L;C:\Database\demo.1].

Experimental animals

Adult *M. cartusiana* snails were collected from infested clover fields in Quftan village, Sumasta district, Beni-Suef Governorate and transported to the laboratory in a plastic bag. Snails were acclimatized by keeping them in plastic containers with moist soil and feeding them lettuce leaves for 14 days.

Laboratory bioaasay

Bait toxicity assay

Different concentrations of clove oil (1.25, 2.5, 5, 10, 20, 50 mg/ml) and thyme oil (1.25, 2.5, 5, 10, 20, 50 mg/ml), were tested as poison baits against *M. cartusiana* and formulated by combining each of these concentrations with 5% molasses + 93% bran. Five grams of bait were placed on the soil surface of each glass box using a plastic sheet. Each tested bait was given to thirty snails (three replicates of ten snails each). Other thirty individuals (three replicates of ten snails each) served as the control group. After 7 days, mortality rates and the LC_{50} value were calculated according to Finney (1971).

Contact toxicity assay

The contact toxicity assay was performed using the method described by Asher and Mirian (1981). In brief, two ml of thyme oil (0.32, 0.625 and 1.25 mg/ml) and clove oil (0.625, 1.25, 2.5 and 5 mg/ml) were distributed on the inner surface of a petri-dish and gently rotated in circles. For each concentration, three replicates were used. Equal numbers of untreated snail replicas were given water and were considered the control group. According to Finney (1971), dead animals were counted daily and the mortality % and LC_{50} were calculated.

Ovicidal activities of the Eos

After the egg deposition, new clutches were retrieved using a fine hair brush. The eggs were divided into 20-egg batches. Each batch of eggs (up to 24 hours old) was placed in a culture dish containing 5 g of sterile moist soil. One ml of LC_{50} concentration of each tested compound was applied directly to egg batches. The control group of eggs received distilled water treatment. Each treatment was assessed in three replicates. The hatchability of the eggs was checked daily for seven days.

Collection and preparation of tissues

The toxicity of EOs against *M. cartusiana* snails was assessed using the topical application approach (contact action) described by Radwan *et al*. (2008). For each treatment, triplicates (each of 10 people) were exposed to the LC_{50} concentrations the thyme and clove oils for 24 hours. The control group received distal water treatment. After removing the snail's shell, the soft tissue was weighed and homogenized in 10 volumes of ice-cold saline solution (w/v ratio) for 60 seconds using a Polytron Kinemetica homogenizer, then centrifuged at 3000 r.p.m. for 15 minutes. Supernatants were isolated and then stored at -80°C until needed.

Biochemical assessment

Oxidant/antioxidant defense biomarker

The lipid peroxidation (MDA) and glutathione (GSH) levels were evaluated adopting the Ohkawa *et al*. (1979) and Beutler *et al*. (1963) methods, respectively, using a reagent kit purchased from Biodiagnostic Company (Egypt).

Metabolic enzyme markers

Alkaline phosphatase activity (ALP) was measured according to the manufacturer's instructions for the Spectrum Diagnostic Kit (ALP, Cat. No. AP 10 20).

Energy reserves biomarker

Total protein (TP) (Cat. No. TP 20 21) was assessed according to Kinsley and Frankel (1939), meanwhile total lipids (TL) (Cat. No. TL 20 10) level were estimated by the methods of Zöllner and Kirsch (1962).

Neurotoxicity marker

Acetylcholinesterase (AChE; EC 3.1.1.7) assay is based on an improved Ellman method, in which thiocholine produced by the action of acetylcholinesterase forms a yellow color with 5,5'-dithiobis (2-nitrobenzoic acid). The intensity of the product color, measured at 412 nm, is proportionate to the enzyme activity in the sample (Ellman *et al*., 1961).

Steroid hormones

Hormone concentrations testosterone (T) and 17 β-estradiol (E) were assayed according to the manufacture instructions of T EIA kit (Enzo Life Science, Michigan, USA, ADI-900-065) and E EIA kit (Cayman Chemical Company, Michigan, USA, item no. 582251).

Histological studies

The digestive gland and ovotestis of the surviving *M. cartusiana* snails were removed and preserved in Bouin's solution for histological studies. According to Mohamed and Saad (1990), these samples were first dehydrated in gradient ethanol, then embedded in paraffin wax and then cut into sections and stained with hematoxylin and eosin. Slides of these organs were examined for any histological changes in comparison to control snails.

Field application

The field efficacy of clove and thyme oil as bait against Monacha snails was compared to the prescribed methomyl compound (2%). At Quftan Village, Sumsta District, Beni-Suef Governorate, twelve clover-growing plots (each 30 m^2) were selected and infested with *M. cartusiana*. For each evaluated compound and the control, three replicates were used. The space between the plots is maintained at least at four metres. The most potent lab-tested concentrations of thyme (50 mg/ml) and clove oils (20 mg/ml) were selected for evaluation against *M. cartusiana* in the field after treatment, live snails were counted in each plot both before and after the application. Henderson and Tilton (Henderson and TILTON, 1955) determined the decline in snail population after 21 days of treatment.

Statistical analysis

Our data was analyzed by one-way analysis of variance (ANOVA) using SPSS (version 20) statistical program (SPSS Inc., Chicago, IL, USA). Analysis of differences between the means of different groups was performed using Duncan's multiple range and two tailed paired t-test. The lethal concentration values and respective 95% confidence limit (CL) of LC_{50} was calculated by Probit analysis (Finney, 1971).

RESULTS AND DISCUSSION

GC Mas analysis of thyme and clove oil

The GC-MS analysis identified the primary components of thyme oil as thymol (33.89%), *p*-cymene (20.48%), carvacrol (4.65%), terpinen-4-ol (3.71%) and linalool (7.74%). For clove oil, the main constituents were eugenol (32.82%), heneicosane (11.32%), acetophenone (11.16%) and benzothiophene-3-carbohydrazide (9.91%) as shown in (Table 1).

Molluscicidial activity of thyme and clove oil against *M. cartusiana* snails

The molluscicidal activity of thyme and clove oils was confirmed through bait and contact toxicity assays (Table 2) with both methods showing dose-dependent effects. In bait toxicity, thyme oil exhibited higher mortality (93.33% at 50 mg, $P<0.001$) compared to clove oil (80.00%, $P<0.05$), with LC_{50} values of 15.45 mg and 16.02 mg, respectively. Contact toxicity assays demonstrated greater

Table 1: Chemical composition of thyme oil and clove oil as detected by GC-MS analysis.

RT%	Compound name of clove oil	Area % (higher than 1%)	RT%	Compound name of thyme oil	Area % (higher than 1%)
16.073	2,4-Decadienal	3.127	6.292	p-cymene	20.485
17.091	Eugenol	32.823	6.29	linalool	3.596
34.268	octadecane	1.078	9.530	endo-borneol	1.776
34.694	Eicosane	6.028	9.78	terpinen-4-ol	3.714
34.840	Heneicosane	11.329	12.38	thymol	33.896
35.203	Tricosane	3.619	12.608	carvacrol	4.653
36.148	Pentacosane	2.671	18.856	caryophyllene oxide	2.237
36.291	Tetracosane	1.017			
36.478	hexacosane	1.450			
40.673	Acetophenone	11.160			
41.456	Benzothiophene-3-carbohydrazide	9.917			

efficacy, with thyme oil causing 100% mortality at 0.625 mg and achieving an LC_{50} of 0.21 mg, compared to clove oil's LC_{50} of 0.548 mg.

Ovicidal activities of the Eos

Both oils showed significant ovicidal activity at LC_{50} concentrations (Fig 1), causing 100% un-hatchability.

Biochemical evaluation

Sub-lethal doses of thyme and clove oils induced oxidative stress, evident from elevated MDA levels and decreased non-enzymatic antioxidant GSH levels in treated snails (Fig 2). Thyme oil caused a greater increase in lipid peroxidation than clove oil (P<0.001).

Table 2: Molluscicidal activity, LC_{50}, LC_{90} and 95% confidence limit (CL) of clove and thyme against *M. cartusiana* snail after 24 h of exposure by Bait and contact toxicity assay.

Dose (mg/ml)	Mortality per cent ± SEM Contact toxicity assay		Dose (mg/ml)	Mortality per cent ± SEM Bait toxicity assay	
	Clove oil	Thyme oil		Thyme oil	Clove oil
50 (5%)	93.33± 4.41^{a}	80.00± 1.67^{a}	2.5(0.25%)	100.00±0.00^{a}	85.00 ± 6.12^{a}
20(2%)	53.33±3.33^{b}	56.67 ± 1.67^{b}	1.25 (0.125%)	100.00±0.00^{a}	82.00 ± 6.04^{a}
10(1%)	46.67±1.66^{b,c}	43.33 ± 3.33^{c}	0.625 (0.063%)	100.00±0.00^{a}	55.00 ± 3.16^{b}
5(0.5%)	40.00±0.00^{c,d}	41.67 ± 4.41^{c}	0.313 (0.0313%)	70.00±3.16^{b}	50.00 ± 2.24^{b}
2.5(0.25%)	33.33±3.33^{d}	26.66± 3.33^{e}	0.156 (0.0156%)	40.00±1.58^{c}	25.00±1.58^{c}
1.25 (0.125%)	16.67±1.67^{e}	21.66 ±1.66^{e}	Control	0.00±0.00^{d}	0.00±0.00^{d}
Control	0.00±0.00^{f}	0.00±0.00^{f}			
LC_{50}(95% confidence limit)	15.45 (8.85-24.44)	16.02 (6.55-27.96)		0.21(0.094-0.282)	0.548 (0.30-1.04)
LC_{90} (95% confidence limit)	44.86 (32.72-77.61)	56.88 (39.70-114.34)		0.342 (0.344-0.730)	2.64(1.72-5.34)

Data represented as mean ± SEM, Superscript of different letters in cells of the same column is significant (P<0.05).

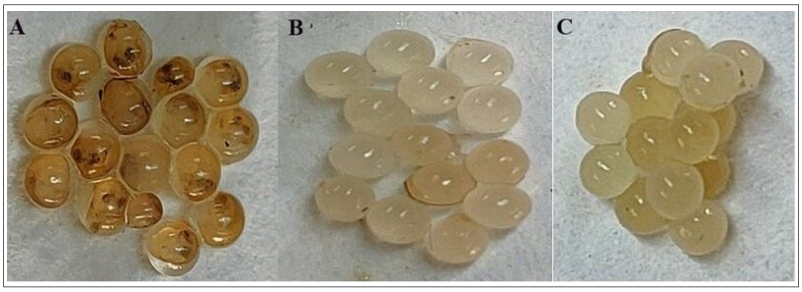
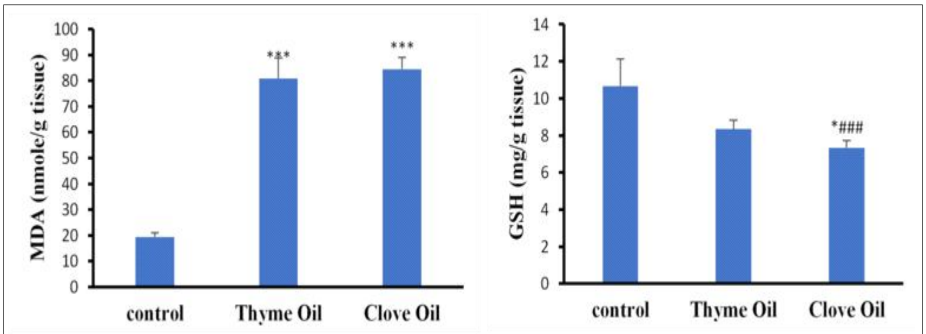


Fig 1: Ovicidal activity of thyme (B) and clove (C) oil against *M. cartusiana* snail after 7 days of LC_{50} exposure in compared to control (A), EM: embryo.

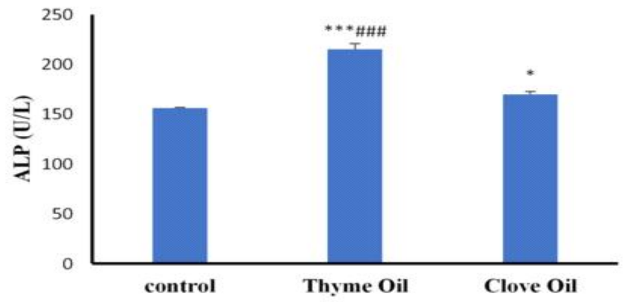


The column represented mean±SEM. statistical significance at P <0.05 using Duncan's multiple range test.
*,*** Significantly different at P <0.05, P <0.001 over their respective controls.
Significantly different at P <0.001 over their thyme treated group.

Fig 2: Oxidant/antioxidant stress biomarkers in *M. cartusiana* exposed to sublethal LC_{50} of thyme and clove oil along with their respective controls.

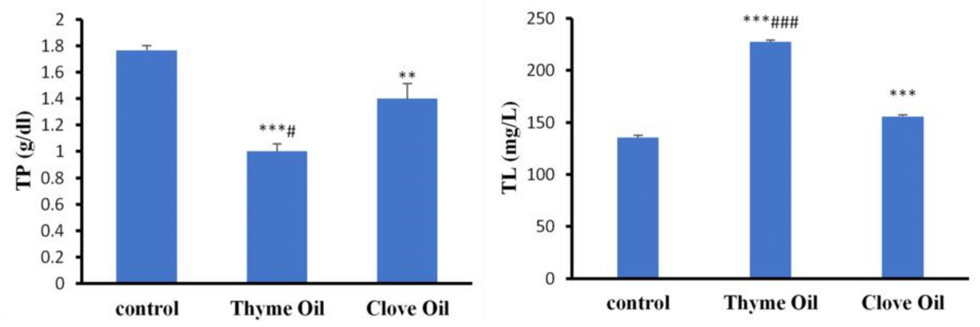
Fig 3 shows that the LC_{50} doses of thyme and clove oil significantly increased ALP activity in snail tissues (*P*<0.001; *P*<0.05, respectively) when compared to control snails. Comparing thyme oil to clove oil, thyme oil significantly increased ALP (*P*<0.001).

Thyme and clove oils significantly increased lipid levels (*P*<0.001) and decreased total protein levels in treated snails (Fig 4). Following exposure to thyme and clove Eos at LC_{50} concentration, levels of T significantly decreased (*P*<0.01, *P*<0.001, respectively). Additionally, when



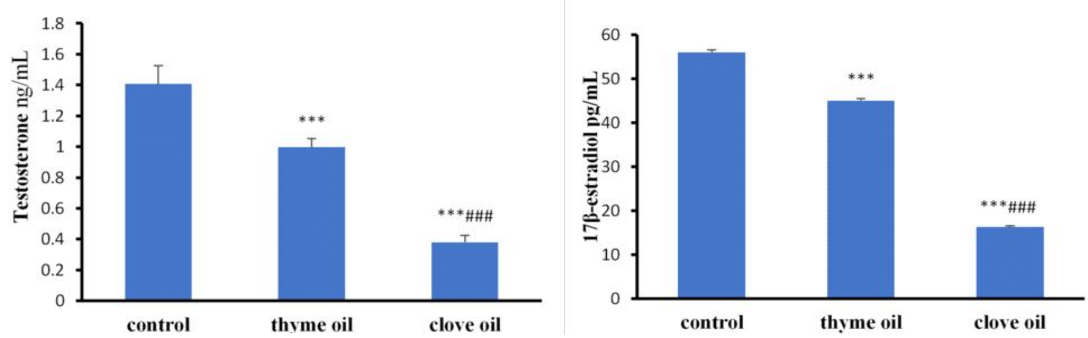
The column represented mean±SEM. statistical significance at *P*<0.05 using Duncan's multiple range test.
*,*** Significantly different at *P*<0.05, *P*<0.001 over their respective controls.
Significantly different at *P*<0.001 over their clove treated group.

Fig 3: Metabolic enzyme markers in *M. cartusiana* exposed to sublethal LC_{50} of thyme and clove oil along with their respective controls.



The column represented mean ± SEM. statistical significance at p≤0.05 using Duncan's multiple range test.
,* Significantly different at *P*<0.01, *P*<0.001over their respective controls.
#,### Significantly different at *P*<0.05, *P*<0.001over their treated group.

Fig 4: Energy reserves biomarker in *M. cartusiana* exposed to sublethal LC_{50} of thyme and clove oil along with their respective controls.



The column represented mean ± SEM. statistical significance at *P*≤0.05 using Duncan's multiple range test.
*** Significantly different at *P*<0.001 over their respective controls.
Significantly different at *P*<0.05 over their thyme treated group.

Fig 5: Steroid hormones in *M. cartusiana* exposed to sublethal LC_{50} of thyme and clove oil along with their respective controls.

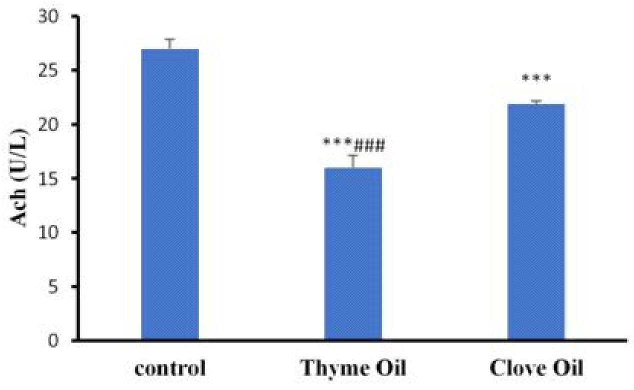
compared to control, both oils had the same effect on the level of E (P<0.001, P<0.001) (Fig 5). Clove oil caused more severely endocrine disruption (P<0.001) than thyme oil did.

Both oils significantly reduced acetylcholine esterase (AChE) activity (Fig 6), with thyme oil exhibiting stronger inhibition P<0.001.

Histopathological evaluation

Histological analysis

The results of the current investigation demonstrated that LC_{50} dose of thyme oil caused rupturing of the germinal epithelium in the digestive gland (Fig 7B), as well as

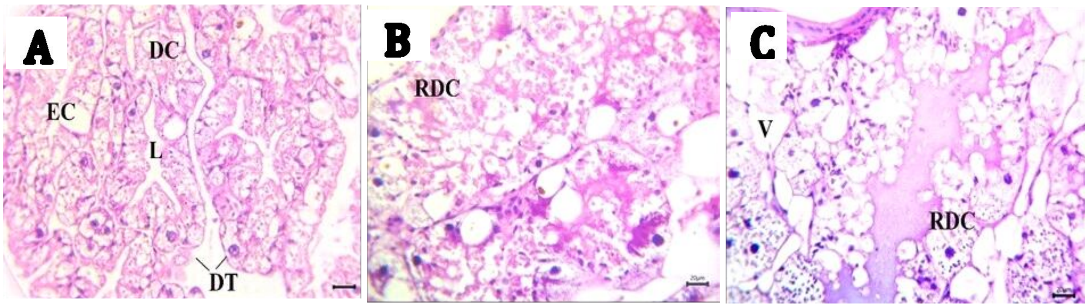


The column represented mean of three replicates ± SE. statistical significance at $P\leq0.05$ using Duncan's multiple range test.

*** Significantly different at P<0.001 over their respective controls.

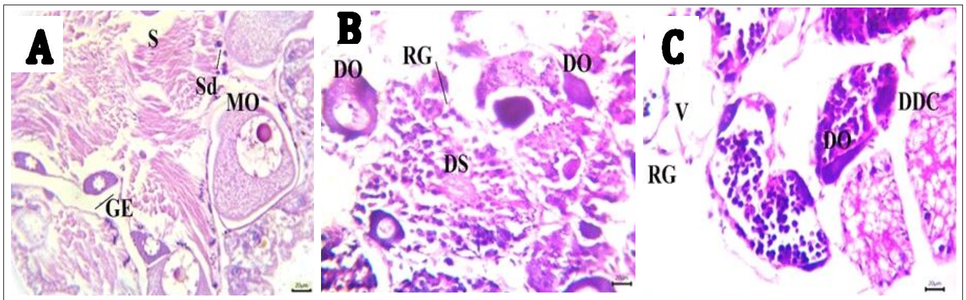
Significantly different at P<0.001 over their clove treated group.

Fig 6: Acetylcholinestrase activity in *M. cartusiana* exposed to sublethal LC_{50} of thyme and clove oil along with their respective controls and treated group.



(A) control, digestive tubule (DT), digestive cell (DC), excretory cell (EC), lumen (L). (B) thyme treated snails showing presence of cellular blebs, rupture (RDC) and degeneration (DDC) digestive cell. (c) clove treated snails showing presence of vacuolation (V), hemocytic infiltration (HI), rupture (RDC) digestive cell (scale bar= 20 µm).

Fig 7: Light micrographs showing effect of thyme and clove oil at LC_{50} on the digestive gland of *M. cartusiana* snails.



(A) control ovotestis, germinal epithelium (GE), sperms (S), spermatide (Sd) and mature ova. (B) thyme treated snails showing deformed sperms (DS), deformed ova (DO (C) clove treated snails showing rupture germinal epithelium (RG), deformed ova (DO), and vacuoles (V).

Fig 8: Light micrographs showing effect of thyme and clove oil at LC_{50} on ovotests of *M. cartusiana* snails.

Table 3: Field performance of thyme, clove and binary mixture against land snails, *M. cartusiana*, comparing with control after 21 days of treatment.

Treatment	No. pretreatment	No. post treatment	t	*P*	Population reduction %
Thyme oil	60.80±7.42	5.60±2.71	5.569	0.005	90.94
Clove oil	68.00±6.32	4.8±1.96	9.914	0.001	93.056
Methomyl	78.40±12.94	5.60±2.40	5.19	0.007	92.97%
Control	96.80±9.67	98.40±7.86	-0.237	0.825	

Data represented as means ± SEM, n=5 (paired t-test P: $P \leq 0.05$).

deformed sperm and ova in the hermaphrodite gland of M. cartusiana snails (Fig 8B). On the other hand, clove oil significantly reduced the amount of sperm and ova along with severe damage (Fig 8C). Additionally, the digestive gland displayed abnormalities such as inflammation, DC degeneration and tubular disturbances with loss of their characteristic form (Fig 7C).

Field efficacy

Field studies demonstrated significant reductions in the *M. cartusiana* population after treatment with thyme and clove oils, achieving reductions of 90.94% and 93.56%, respectively Table 3.

The chemical compositions of thyme and clove oils in this study are consistent with those reported by Haro-Gonzalez *et al.* (2021) and Pandey *et al.* (2022), though slight variations are expected due to factors like geographic origin, cultivation conditions and extraction techniques (Pavela and Benelli, 2016). The phenolic compounds thymol and eugenol are recognized for their potent bioactivity and are likely responsible for the strong molluscicidal effects observed (Radwan and Gad, 2021).

The high mortality rates in both bait and contact assays confirm the potent molluscicidal properties of thyme and clove oils, with thyme oil showing slightly greater efficacy. These findings align with earlier work by El-Zemity and Radwan (2001) and Abdel-Rahman and Amal (2020), reinforcing the topical effectiveness of essential oils against snails.

The ovicidal activity observed supports the hypothesis that essential oils interfere with embryonic development, as previously reported by Ali (2017). This may be attributed to disruption of hormonal regulation or direct damage to egg membranes.

Biochemical assays indicated that sub-lethal doses of both oils induce oxidative stress, evidenced by increased MDA and decreased GSH levels. These results echo findings by Lazarevic *et al.* (2020) and Gad *et al.* (2023), who observed similar oxidative responses in invertebrates exposed to essential oils.

Increased ALP activity, especially with thyme oil, suggests lysosomal membrane destabilization and cellular damage, consistent with prior observations (Shekari *et al.,* 2008; Mobarak *et al*., 2015). The accompanying increase in lipid levels and reduction in protein content may be due to stress-induced catabolism and impaired metabolic functions, in line with Capowiez *et al*. (2015).

The endocrine-disrupting effects, particularly the reduction in testosterone and estradiol levels, indicate interference with steroidogenesis, supporting conclusions by Desouky *et al*. (2022). The more severe hormonal disruption caused by clove oil implies differential activity of its constituents on endocrine pathways.

AChE inhibition by both oils, especially thyme oil, suggests neurotoxicity. This is likely due to thymol and eugenol, known AChE inhibitors (Liao *et al*., 2016; El Gohary *et al*., 2021). Such inhibition can lead to overstimulation of the nervous system and ultimately, death.

Histopathological changes observed in the digestive and hermaphrodite glands reinforce the biochemical and hormonal findings. The degeneration of reproductive tissues indicates a significant impact on fertility, as similarly reported by Desouky *et al*. (2022) and Hategekimana and Erler (2020).

Finally, the field efficacy results highlight the practical potential of these essential oils as eco-friendly molluscicidal agents. The comparable performance to methomyl suggests their possible integration into integrated pest management strategies, as supported by previous field studies (Ismail and AbdEl-Kader, 2011; Powell and Bowen, 1996).

CONCLUSION

Our research highlights the potential of thyme and clove oil as a biorational molluscicide against land snails as an alternative for the chemical managements.

ACKNOWLEDGEMENT

This work was supported by Ongoing Research Funding Program (ORF-2025-03), King Saud University, Riyadh, Saudi Arabia.

Authors contributions

Conceptualization; Heba Abdel Tawab, Ebtesam A. Yousef, Heba Y. Ahmed; Data curation; Heba Abdel Tawab, Heba Y. Ahmed, Abdel-Baki, A.A.S., Ahmed O Hassan; Formal analysis; Heba Abdel Tawab, Heba Y. Ahmed; Funding acquisition; Abdel-Baki, A.A.S., Saleh Al-Quraishy; Investigation; Heba Abdel Tawab, Ebtesam A. Yousef; Methodology Heba Abdel Tawab, Ebtesam A. Yousef, Heba Y. Ahmed Supervision; Shawky M Aboelhadid, Abdel-Baki, A.A.S.; Validation; Saleh Al-Quraishy, Shawky M. Aboelhadid, Ahmed O Hassan; Visualization; Heba Abdel Tawab; Heba Y. Ahmed Roles/Writing - original draft; Heba Abdel Tawab, Writing - review and editing; Abdel-Baki, A.A.S., Shawky M. Aboelhadid.

Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Ethics approval

Not applicable.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships.

REFERENCES

Abd El-Halim, S.M., Ali, I.H.H., El-Sayed, S.A.M., Ali, R.F. (2021). Preliminary study on survey and population dynamic of the Terrestrial Snail; *Monacha obstructa* (Pfeiffer) (Hygromiidae, Mollusca) at Crop Fields in Fayoum Governorate, Egypt. *Pakistan Journal of Biological Sciences*. **24(9):** 928-938. doi: 10.3923/pjbs.2021.928.938

Abdel-Halim, K.Y., Aly, G.F., Seloma, A.O., Abdou, G.Y. (2021). Potential toxic effects of nano-derived form to thioben carb on two land snails, *Theba pisana* and *Eobania vermiculata*. *Egyption Journal of Chemistry.* **64(12):** 7539-7549. https://doi.org/10.21608/ejchem.2021.94585.4447.

Abdel-Rahman, A.H.E. and Amal, H.E. (2020). Usage of some botanical oils to control the land snail *Monacha* sp. (Gastropoda: Helicidae). *Egyptian Journal of Plant Protection Research Institute*. **3:** 1241-1254.

Ali, M.A. (2017). Comparison among the toxicity of thymol and certain pesticides on adults survival and egg hatchability of the glassy clover snail *Monacha cartusiana* (MÜLLER). *Journal of Plant Protection and Pathology, Mansoura University*. **8(4):** 189-194.

Aljabeili, H.S., Barakat, H., Abdel-Rahman, H.A. (2018). Chemical composition, antibacterial and antioxidant activities of thyme essential oil (*Thymus vulgaris*). *Food and Nutrition Sciences*. **9:** 433-446. doi: 10.4236/fns.2018.95034.

Asher, R.S. and Mirian, F. (1981). The residual contact toxicity of Bay Sir 8514 to *Spodopteralittoralis* larva. *Phytoparasitica*. **9(2) (1981):** 133-137

Barker, G.M. (2002). Gastropods as pests in New Zealand pastoral agriculture, with emphasis on Agriolimacidae, Arionidae and Milacidae. In Barker, G.M. Molluscs as crop pests CABI. pp. 361-423. ISBN 0-85199-320-6.

Barua, A., Roy, S., Handique, G., Bora, F.R., Rahman, A., Muraleedharan, P.D.N. (2017). Clove oil efficacy on the red spider mite, *Oligonychus coffeae* Nietner (Acari: Tetranychidae) infesting tea plants. *Proceedings of the Zoological Society India*. **70:** 92-96. https://doi.org/10.1007/s12595-015-0147-6.

Beutler, E., Duron, O., Kelly, B.M. (1963). Improved method for the determination of blood glutathione. *Journal of Laboratory and Clinical Medicine*. **61:** 882-888.

Bozin B., Mimica-Dukic N., Simin N., Anackov G. (2006). Characterization of the volatile composition of essential oils of some lamiaceae spices and the antimicrobial and antioxidant activities of the entire oils. *Journal of Agricultural and Food Chemistry*. **54:** 1822-1828. doi: 10.1021/jf051922u.

Capowiez, Y., Bottinelli, N., Sammartino, S., Michel, E., Jouquet, P. (2015). Morphological and functional characterisation of the burrow systems of six earthworm species (*Lumbricidae*). *Biol. Fertil. Soils.* **51:** 869-877.

Capowiez, Y., Rault, M., Mazzia, C., Belzunces, L. (2003). Earthworm Behaviour as a Biomarker - A Case Study Using Imidacloprid: The 7th International Symposium on Earthworm Ecology. Cardiff. Wales. 2002. *Pedobiologia*. **47:** 542-547.

Da Silva, F.F.M., Monte, F.J.Q., de Lemos, T.L.G., do Nascimento, P.G.G., de Medeiros Costa, A.K., de Paiva, L.M.M. (2018). Eugenol derivatives: Synthesis, characterization and evaluation of antibacterial and antioxidant activities. *Chemistry Center Journal*. **12(1):** 34. Published 2018 Apr 3. doi:10.1186/s13065-018-0407-4.

Dassanayake, M.K., Chong, C.H., Khoo, T-J., Figiel, A., Szumny, A., Choo, C.M. (2021). Synergistic field crop pest management properties of plant-derived essential oils in combination with synthetic pesticides and bioactive molecules: A review. *Foods*. **10(9):** 2016. https://doi.org/10.3390/foods 10092016.

Desouky, M.M.A., Abd El-Atti, M.S., Elsheakh, A.A., Elgohary, W.S. (2022). Effect of *Eucalyptus globulus* oil and *Ricinus communis* methanolic extract as potential natural molluscicides on the reproductive biology and some antioxidant enzymes of the land snail, *Theba pisana*. *Heliyon.* **20: 8(12):** e12405. doi: 10.1016/j.heliyon.2022.e12405.

El Gohary, E.E., Farag, S.M., El-Sayed, A.A., Khattab, R.R., Mahmoud, D.M. (2021). Insecticidal activity and biochemical study of the clove oil (*Syzygium aromaticum*) nano-formulation on *Culex pipiens* L. (Diptera: Culicidae). *Egyptian Journal of Aquatic Biology and Fisheries*. **25:** 227-239.

Ellman, G.L., Courtney, K.D. andres, V.J. Feather-Stone, R.M. (1961). A new and rapid colorimetric determination of acetylcholinesterase activity. *Biochemical Pharmacology.* **7:** 88-95. doi: 10.1016/0006-2952(61)90145-9.

El-Zemity, S.R. and Radwan, M.A. (2001). Molluscicidal and antifeedant activity of some essential oils and their major chemical constituents against Theba pisana snails. *Arab Universities Journal of Agricultural Sciences*. **9:** 483-493.

Finney, D.J. (1971). Probit Analysis, 3rd edn. Cambridge University Press, Cambridge, UK. Godan, D., 1983. Pest Slugs and Snails, Biology and Control. Springer-Verlag, Berlin/ New York, p. 443.

FOOD, D. ADMINISTRATION and U. FDA (2007). Guidance for Industry: Concerns related to the use of closve oil as anesthetic for fish. 2007, Rockville.

Gad, A.F., Abdelgalil, G.M., Radwan, M.A. (2023). Bio-molluscicidal potential and biochemical mechanisms of clove oil and its main component eugenol against the land snail, *Theba pisana*. *Pesticide Biochemistry and Physiology.* **192:** 105407. doi: 10.1016/j.pestbp..10540.

Gavaric, N., Mozina, S.S., Kladar, N., Bozin, B. (2015). Chemical profile, antioxidant and antibacterial activity of thyme and oregano essential oils, thymol and carvacrol and their possible synergism. *Journal of Essential Oil Bearing Plants.* **18:** 1013-1021. https://doi.org/10.1080/0972060X.2014.971069.

Godan, D. (1983). Pest Slugs and Snails: Biology and Control. Springer-Verlag, Berlin. 445 pp.

Haro-Gonzalez, J.N., Castillo-Herrera, G.A., Martínez-Velazquez, M., Espinosa-Andrews, H. (2021). Clove essential oil [*Syzygium aromaticum* (L.) Myrtaceae]: Extraction, chemical composition, food applications and essential bioactivity for human health. *Molecules*. **26:** 6387. doi: 10.3390/molecules26216387.

Hategekimana, A. and Erler, F. (2020). Fecundity and fertility inhibition effects of some plant essential oils and their major components against *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae). *Journal of Plant Diseases and Protection*. **127:** 615-623.

Ismail, S.A.A. and Abdel Kader, S.M. (2011). Clove: Is it has a molluscicidal activity against land snails (*Monacha cartusiana*)? *Journal of Plant Protection and Pathology*. **2:** 561-569.

Isman, M.B. (2000). Plant essential oils for pest and disease management. *Crop Protection.* **19:** 603-660.

Kinsley, S.R. and Frankel, S.J. (1939). The determination of serum total protein and albumin- globulin ratio. *Journal of Biological Chemistry.* **128:** 131-137.

Klein, M.L., Chastain, T.G., Garbacik, C.J., Qian, Y.L., Mc Donnell, R.J. (2020). Acute toxicity of essential oils to the pest slug *Deroceras reticulatum* in laboratory and greenhouse bioassays. *Journal of Pest Science.* **93:** 415-425. doi: 10. 1007/s10340-019-01154-0.

Lazarevic, J., Jevremovi, S., Kostic, I., Kostic, M., Vuleta, A., Jovanovi, S.M., Jovanovic, Š.D. (**2020).** Toxic, oviposition deterrent and oxidative stress effects of *Thymus vulgaris* Essential Oil against *Acanthoscelides obtectus*. *Insects*. **11(9):** 563. https://doi.org/10.3390/insects11090563.

Liao, M., Xiao, J-J., Zhou, L-J., Liu, Y., Wu, X-W., Hua, R-M., Wang, G-R., Cao, H-Q. (2016). Insecticidal activity of *Melaleuca alternifolia* essential oil and RNA-Seq analysis of *Sitophilus zeamais* transcriptome in response to oil fumigation. *PLoS ONE*. **11:** e0167748.

Mobarak, S.A., Kandil, R.A., Abd ElKader, S.M. (2015). Effect of clove plant extract on land slug and their reproductive system under laboratory and field conditions. *Journal of American Science*. **11(12):** 250-255. doi: 10.7537/marsjas111215.32.

Mohamed, S.H. and Saad, A.A. (1990). Histological studies on the Hermaphrodite gland of *Limnaea caillaudi* and *Biomphalaria alexandrina* upon infection with certain larval trematodes. *Egyption Journal of Histology*. **13(1):** 47-53.

Muhie, S.H. (2022). Novel approaches and practices to sustainable agriculture. *Journal of Agriculture and Food Research*. **10(2):** 10044. doi: 10.1016/j.jafr.2022.100446

Murshed, M., Al Ansi, A.N. and Mohamed, B.O. (2024). Potential of Animal Bile as Bio-molluscicide against Land Snail *Monacha cartusiana*. *Indian Journal of Animal Research*. **58(7):** 1203-1208. doi: 10.18805/IJAR.BF-1790.

Nasiou, E. and Giannakou, I.O. (2020). The potential of eugenol as a nematicidal agent against *Meloidogyne javanica* (Treub) Chitwood. *Journal of Nematology*. **52:** e2020-e2103. doi: 10. 21307/jofnem-2020-103.

Ohkawa, H., Ohishi, N., Yagi, K. (1979). Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. *Analytical Biochemistry*. **95:** 351-358. doi: 10.1016/0003-2697(79) 90738-3.

Pandey, V.K., Shams, R. Singh, R. Dar, A.H. Pandiselvam, R. Rusu, A.V. Trif, M. (2022). A comprehensive review on clove (*Caryophyllus aromaticus* L.) essential oil and its significance in the formulation of edible coatings for potential food applications. *Frontier in Nutration.* **9:** 987674. https://doi.org/ 10.3389/fnut.2022.987674.

Pavela, R. and Benelli, G. (2016). Essential oils as ecofriendly biopesticides? Challenges and constraints. *Trends in Plant Science*. **21:** 1000-1007. doi: 10.1016/j.tplants.2016.10.005.

Pathak, V.M., Verma, V.K., Rawat, B.S., Kaur, B., Babu, N., Sharma, A., Dewali, S., Yadav, M., Kumari, R., Singh, S., Mohapatra, A., Pandey, V., Rana, N., Cunill, J.M. (2022). Current status of pesticide effects on environment, human health and it's eco-friendly management as bioremediation: A comprehensive review. *Frontier in Microbiology.* **17(13):** 962619. doi: 10.3389/fmicb.2022.962619.

Powell, A.L. and Bowen, I.D. (1996). The screening of naturally-occurring compounds for use as seed treatments for the protection of wheat against slug damage. IN: Henderson IF (ed) Slug and Snail Pests in agriculture. BCPC Proceedings No. 66. BCPC, Farnham, UK, pp 231-236

Radwan, M.A. and Gad, A.F. (2021). Essential oils and their components as promising approach for gastropod mollusc control: A review. *Journal of Plant Diseases and Protection*. **128:** 923-949.

Radwan, M.A., EL-Zemity, S.R., Mohamed, S.A., Sherby, S.M. (2008). Larvicidal activity of some essential oils, monoterpenoids and their corresponding N-methyl carbamate derivatives against *Culex pipiens* (Diptera: Culicidae). *International Journal of Tropical Insect Science.* **28:** 61-68.

Rana, I.S., Rana, A.S., Rajak, R.C. (2011). Evaluation of antifungal activity in essential oil of the *Syzygium aromaticum* (L.) by extraction, purification and analysis of its main component eugenol. *Brazilian Journal of Microbiology*. **42:** 1269-1277. doi: 10.1590/S1517-83822011000400004.

Sekar, G. C., Ibohal, K., R., Loganathan and Okendro, S. (2025). Eco-friendly approach for the management of rust red flour beetle, *Tribolium castaneum* (Herbst) on Wheat Grains. *Agricultural Science Digest.* **45(1):** 144-147. doi: 10. 18805/ag.D-5454.

Shekari, M., Sendi, J.J., Etebari, K., Zibaee, A., Shadparvar, A. (2008). Effects of *Artemisia annua* L. (Asteracea) on nutritional physiology and enzyme activities of elm leaf beetle, *Xanthogaleruca luteola* Mull. (Coleoptera: Chrysomellidae). *Pesticide Biochemistry and Physiology.* **91:** 66-74. https:// doi.org/10.1016/j.pestbp.2008.01.003.

Simms, L.C., Mullins, C.E., Wilson, M.J. (2002). Seed dressings to control slug damage in oilseed rape. *Pest Managment Science*. **58:** 687-694. doi: 10.1002/ps.514.

Sujatha, K., Ramamoorthy, K. and Sivasubramaniam, K. (2015). Efficacy of some indigenous plant products against pulse beetle *Callosobruchus spp* on cowpea (Vigna unguiculata L. Hepper.) seeds. *Agricultural Science Digest*. **35(1):** 74-77. doi: 10.5958/0976-0547.2015.00016.6.

Yazdani, E., Jalali-Sendi, J., Hajizadeh, J. (2014). Effect of *Thymus vulgaris* L. and *Origanum vulgare* L. essential oils on toxicity, food consumption and biochemical properties of lesser mulberry pyralid Glyphodes pyloalis Walker (Lepidoptera: Pyralidae). *Journal of Plant Protection Research*. **54:** 53-61. doi: https://doi.org/10.2478/jppr-2014-0008.

Zöllner, N. and Kirsch, K. (1962). Colorimetric method for determination of total lipids. *Journal of Experimental Medicine*. **135:** 545-550.http://dx.doi.org/10.1007/BF02045455.