

MOLLUSCICIDAL AND ANTIFEEDANT IMPACTS OF SOME WEED EXTRACTS ON THE LAND SNAIL *MONACHA OBSTRUCTA* (PFEIFFER)

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ABSTRACT

The molluscicidal and antifeedant activity of three weeds extracts namely primpernel (*Anagallis arvensis* L.) (Primulaceae), nightshade (*Solanum nigrum* L.) (Solanaceae) and nutsedge (*Cyperus longus* L.) (Cyperaceae) were evaluated against the adult stage of *Monacha obstructa* (Pfeiffer, 1842) under laboratory conditions. Three methods of bioassay were used i.e. bait, residual film (contact) and antifeeding techniques. The results indicated that chloroform and hexane extracts of primpernel appeared highly toxic when applied as baits on *M. obstructa* adults while nutsedge was found to be lower toxicity based on LC₅₀ values. Also, chloroform extract of nightshade and hexane extract of primpernel were the most potent with contact whereas nutsedge was the least effective. On the other hand, of either chloroform or hexane extract primpernel tended to be more effective as antifeedant compound than nightshade. In contrast, nutsedge chloroform extract revealed no action as a feeding deterrent whereas the same hexane extract was effective based on SC₅₀ levels. Moreover, fifteen compounds were identified by GC/MS analysis in the chloroform extract of primpernel. Palmatic acid methyl ester (fatty acid) was the dominant compound representing 25.8% followed by aromadendrene (sesquiterpene) forming 15.2% and 2 – tertbutyl - 4- isopropyl – 5- methylphenol (phenolic compound) having 13.9%. In addition to the presence of two steroids, two dicarboxylic acids and only one ketone compound was identified. Finally, the salt of carbamic acid (ammonium carbamate) and the ester of phosphoric acid (tributyl phosphate) were also detected. This study reveals that chemical compounds present in chloroform extract of primpernel has effective source as a molluscicide extracted of plant origin and has potential beneficial control against *M. obstructa* snails.

Keywords: Weed extracts, Toxicity, Antifeeding effect, *Monacha obstructa*, Land snail, Molluscicides, Identification, GC/MS analysis.

INTRODUCTION

Gastropods including terrestrial snails are the second most diverse animal group after arthropods and other invertebrates. Terrestrial snails are considered very serious pests resulting a significant loss of several agricultural crops causing a severe damage to fruit trees as well as to field crops, vegetables and ornamental plants in many countries.

In Egypt, these pests are widespread in Delta region with excessive abundant in North Coast belt of Mediterranean Sea. (Kassab and Daoud, 1964; El-Okda, *et al.*1977; El-Okda, 1980; Nakhla and Tadros 1995; Mahmoud and Awad 2008) and

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other countries around the world (Godan, 1983; Frank, 1998; El-Zamity, 2006; Barker, 2008; Davis *et al.* 2010). Currently, many growers are aware of the heavy loss caused by terrestrial molluscs their importance in Egyptian agricultural fields that became epidemic pests in many governorates i.e. Alexandria, El Sharkia, El Behiera, Giza, EL Qaluiobia and Fayoum governorates. (Nakhla *et al.* 2002; Abd El Wahab, 2004; Ali, 2006; Mohamed and Ali, 2011a; 2011b).

The plant kingdom is one of the rich sources for pest control. Active substances extracted from plants are considered naturally alternatives to find new, safe and effective chemicals act as antifeedants, repellents, synergists and toxicants. Synthetic pesticides lead to environmental pollution and pest resistance with their effects on the beneficial living organisms and phytotoxicity. The toxic effect of plant extracts against the land snail *Monacha obstructa* was reported by other researchers (Speiser *et al.* 1992; Ghamry 1997; Dodds and Hendrson 2002; Ebenso 2004; Shabad 2010; Anjali *et al.* 2014; Ghaly 2016 and Ding *et al.* 2018). The objective of the present investigation is to study the molluscicidal effect of three weed extracts namely primpernel, nightshade and nutsedge against the adult stage of land snail *M. obstructa* as well as identification of chemical components of the more active extract using GC/MS analysis.

MATERIALS AND METHODS

The terrestrial snails *Monacha obstructa* were collected from Tamiya town in Fayoum Governorate (26,488°29' N 30°58.529' E) (in November and December 2018) where more humid and low temperature degree as activity season for terrestrial gastropods. The preferred hosts were Egyptian clover (*Trifolium alexandrinum* L.) (Fabaceae) and on wild weeds i.e. Chicory (*Cichorium pumilum* Jacq.) and annual Sowthistle *Sonchus oleraceus* L. that both belong to (Asteraceae) (Approximately the experiment implemented in February to April 2018 as activity season to the snails. primpernel (*Anagallis arvensis* L.) was collected from the same location of Tamiya town in Fayoum Governorate, while nightshade (*Solanum nigrum* L.) (Solanaceae) and nutsedge (*Cyperus longus* L.) (Cyperaceae) were collected from the agricultural station, Fayoum University in Dar Ramad district. The collected samples were transferred to the laboratory Fayoum University of Agriculture Zoology department of plant protection and starvation for 24 hours.

• **Weed extracts:**

Weeds selected for this study were nutsedge, *Cyperus longus* L. (perennial-cyperaceae); primpernel, *Anagallis arvensis* L. (annual winter weed –Primulaceae) and nightshade, *Solanum nigrum* L. (annual summer weed-Solanaceae). Rhizomes of the former weed whereas leaves and fruits of the two latter were utilized. Samples of 100 g each were in a grinder and extracted using either; chloroform or hexane, at the rate of 2 ml/g plant material as described by Su and Horvat (1981). After 24 hours, the extract was filtered through anhydrous sodium sulphate and evaporated to dryness under vacuum using a rotary evaporator in a water bath at 50°C. The crude extract

was then weighed and adjusted to 10 ml volume with acetone and Kept in a refrigerator until being tested.

- **Molluscicidal activity of weed extracts:**

Three different tests were carried out in laboratory using three weed extracts assayed on the land snail *Monacha obstructa* adult.

- **Poison bait test:**

The poison baits were prepared according to Ebenso (2004). This bait was designed to achieve the following components: 1 ml toxicant + 5ml molasses + 8g bran + 15 ml water. Samples of 5 g of this bait were placed on plastic sheet on the soil surface in each appropriate box (10 x 3). Four serial concentrations from each extract were used (0.25, 0.50, 1.0, 2.0 g/ml) for both the solvents. Each concentration was replicated four times and each replicate contained ten adults, which exposed to the candidate concentration of the tested weed extract. Control treatment of 40 adults was applied at the same technique without toxicant was added. Mortality percentages were calculated after 24, 48 and 96 hours of exposure and corrected by using Abbott formula (1925) as follows:

$$\% \text{ corrected mortality} = \frac{\text{observed mortality} - \text{control mortality}}{100 - \text{control mortality}} \times 100$$

- **Residual film (contact) test:**

A modification of the technique used by Ascher and Mirian (1981) on the adult of *M. obstructa* was adopted. For each extract tested, one ml dilution in acetone was evenly spread inside a petridish, 9 cm dia., by moving the dish gently in circles. The acetone evaporates leaving a thin film of the crude extract. Petridishes used as control was treated with one ml acetone only. Ten adults of *M. obstructa* average weight 23 mg/adult were exposed to this thin film for 24 hrs in each of four replicate dishes. Mortality counts were recorded 24 hrs after treatment and the obtained results were corrected for the natural mortality using Abbott formula as mentioned before (concentrations should be mentioned here as well).

- **Antifeeding effect test:**

Plant discs of lettuce leaves, 7 cm diameter, placed each in a petridish, 9 cm diameter. The petridish was covered with muslin and closed carefully with rubber bands to prevent snails from escaping, were treated with the tested concentrations (Hexane or chloroform with accurate concentrations) of the weed extracts, by the use of a pipette (mentioned the volume according to each concentrations). Ten-weighted *M. obstructa* adult were placed on each disc; in four replications per concentration. Adults were weighed after 24 hrs, and control discs were treated with acetone alone. Another control treatment was set up by complete starvation of 40 adults throughout the test period. The antifeeding effect was measured as starvation percentage and was corrected according to the following formula (Moustafa, 1969).

$$\text{Percent starvation} = \frac{C - E}{C - S} \times 100$$

Where:

E = mean weight gain of treated adult

C = mean weight gain of fed control adult

S = mean weight gain of unfed control adult

Toxicity lines:

Stock concentrations in acetone (W/V) of each of the three weed extracts used in this study were prepared and kept under refrigeration in glass stoppered bottles. Serial dilutions were made such that each was half the strength of the preceding one. Four concentrations per tested material 0.25, 0.50, 1.0 and 2.0 g/ml were applied either as poisonous baits or residual film (contact) whereas 0.1, 0.3, 0.6 and 1.2 g/ml were used for antifeeding effect to draw the Ld-p lines for toxicity tests with at least four replicates for each concentration.

- **Identification by gas chromatography-mass spectrometry (GC/MS) analysis**

The GC column was a 30 m (0.25 mm i.d., film thickness 0.25 Mm) HP-SMS (5% diphenyl) dimethylpolysiloxane capillary column. The GC conditions were as follows; injector temperature, 240° C; column temperature, isothermal at 50° C for 2 min, then programmed to 280° C at 6° C/ min and held at this temperature for 2 min; ion source temperature, 200° C; detector temperature, 300° C. Helium was used as the carrier gas at the rate of 1 ml/min. The effluent of the GC column was introduced directly into the ion source of the MS. Spectra were obtained in the EI mode with 70 eV ionization energy. The sector mass analyser was set to scan from 40 to 400 amu for 5 s. These data were obtained from environmental and food pollutants laboratory at Faculty of Agriculture, Fayoum University.

- **Statistical analysis:**

Toxicological data obtained were subject to statistical analysis to evaluate the relative efficiency of the tested molluscicides. Mortality curves (Ld-p lines) were drawn using Probit analysis program according to the method developed by Finney (1971) to estimate the concentration required to kill 50 and 90 % (LC₅₀ and LC₉₀) in addition to the concentration which caused 50 and 90 % starvation (SC₅₀ and SC₉₀). The value of slope ± S E was also determined.

RESULTS AND DISCUSSION

- **Toxicity of three weed extracts against the adult stage of *M. obstructa***

The effect of three weeds extracts namely: primpernel, nightshade and nutsedge as baits, residual film and antifeedants using chloroform and hexane solvents were tested.

- **Weed extracts as baits:**

- **A.1. Chloroform extracts:**

Data in table (1) indicated that primpernel extract at all different periods of exposure as bait proved to be the highly toxic with LC₅₀ values of 1.07, 0.85 and 0.71

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g/ml after 24, 48 and 96 hrs of treatment respectively. In contrast, nutsedge extract exhibited the lower toxicity with LC₅₀ of 3.09 g/ml after 24 hrs and 1.66 g/ml after 96 hrs unless nightshade was the last extract with LC₅₀ of 2.62 g/ml after 48 hrs. The relative potency at the LC₅₀ values elucidated that primpernel was 2.89 times more toxic than nutsedge after 24 hrs reduced to 2.34 times after 96 hrs. Therefore, the toxicity index for nutsedge extract was 34.6 % as effective as primpernel after 24 hrs increased to 42.7% after 96 hrs exposures. In general, as the exposure period to the bait was extended from 24 to 96 hrs, the toxicity on *M. obstructa* adult was increased and the value of LC₅₀ was decreased.

A.2. Hexane extracts:

It is clear from table (2) that primpernel extract was the most toxic based on LC₅₀ value followed by nightshade extract while nutsedge extract was the least effective to the adult stage *M. obstructa*. The LC₅₀ values after 24 hrs exposures were 2.05 and 2.34 g/ml for primpernel and nightshade extracts respectively. Conversely, the adults of *M. obstructa* were not affected by nutsedge extract through this period to the poison bait. The LC₅₀ values after 96 hrs exposures were decreased to 1.22 g/ml for primpernel and 1.50 g/ml for nightshade whereas nutsedge showed slight toxicity on *M. obstructa* with LC₅₀ was 5.05g/ml. The relative potencies at the LC₅₀ values for primpernel was 4.88 times more potent than nutsedge after 48 hrs declined to 4.14 times after 96 hrs. Consequently, the toxicity index by nutsedge was 20.5 % as toxic as primpernel after 48 hrs enhanced to 24.2% after 96 hrs exposure.

• Weed extracts as contact poisons:

Data presented in table (3) revealed that chloroform extract of nightshade and hexane extract of primpernel were the most effective with LC₅₀ of 0.93 and 1.13 g/ml respectively whereas nutsedge was the least toxic with LC₅₀ of 1.24 and 1.97 g/ml in both extracts. The relative potencies at the LC₅₀ values showed that chloroform extract of nightshade and hexane extract of primpernel were 1.33 and 1.74 times as toxic as nutsedge. Thus, the toxicity index at the same values illustrated that chloroform and hexane extracts of nutsedge were 75.0 and 57.4 % as effective as nightshade by chloroform and primpernel by hexane. Toxic action of plant extracts as molluscicides on land snails was also reported by Ghamry (1997) using *Anagallis arvensis* extract; by Ebenso (2004) using *Azadirachta indica* extract; by Abd El-Halim (2007) using *Foeniculum vulgare l.* extract; by Shabad (2010) using *Artemisia annua* extract; by Anjali *et al.* (2014) using *Solanum nigrum* extract and by Ding *et al.* (2018) using *Ambrosia artemisiifolia* extract.

• Weed extracts as antifeedants:

Data in table (4) indicate that primpernel extract either chloroform or hexane was more effective than nightshade based on SC₅₀. Nutsedge chloroform extract was not effective as antifeedant on the contrary to the same extract by hexane that exerted antifeeding effect with less potent compared to the other tested extracts. Consequently, the SC₅₀ levels for primpernel were 0.22 g/ml by chloroform and 0.78 g/ml by hexane. Also, these levels of SC₅₀ for nightshade were 2.78 g/ml by

chloroform and 1.15 g/ml by hexane while nutsedge by hexane was the least active with SC₅₀ were 4.16 g/ml. Therefore, chloroform and hexane extracts of primpernel exhibited 12.6 and 5.33 folds more potent than nightshade by chloroform and nutsedge by hexane respectively. Plant extracts that acted as antifeedants against molluscs was recorded on *Nerium oldeander* by Coto and Saunders (1987); on *Adenostyles alliariae* by Speiser *et al.* (1992); on *Ammi majus* by Zedan *et al* (2001); on *Conium maculatum* by Dodds and Hendrson (2002) and on *Lupinus termis* by Azzam *et al* (2014) whom found that these molluscicidal plants showed a positive antifeeding impacts.

Table (1): LC₅₀, LC₉₀, Slope, toxicity index and relative potency values for chloroform weed extracts at different exposure periods against the adult stage of *Monacha obstructa*. (as baits)

Weed extracts	LC ₅₀ g/ml	95% confidence limits		LC ₉₀ g/ml	SE ±Slope	Toxicity index at		Relative potency at	
		Lower	Upper			LC ₅₀	LC ₉₀	LC ₅₀	LC ₉₀
hours 24After									
Primpernel	1.07	0.74	1.52	3.64	0.58±2.40	100	100	2.89	5.03
Nightshade	2.75	1.79	367	9.70	1.07±2.34	39.0	37.5	1.12	1.89
Nutsedge	3.09	1.72	484	18.3	0.73±1.66	34.6	20.0	1.00	1.00
hours 48After									
Primpernel	0.85	0.60	1.20	3.57	0.43±2.06	100	100	3.08	3.22
Nightshade	2.62	1.63	51.9	11.5	0.84±1.99	32.4	31.0	1.00	1.00
Nutsedge	2.06	1.27	16.6	11.2	0.72±1.75	41.2	31.9	1.27	1.03
hours 96After									
Primpernel	0.71	0.47	1.00	3.00	0.46±2.05	100	100	2.34	2.42
Nightshade	1.45	0.92	2.58	5.66	0.69±2.16	48.9	53.0	1.14	1.28
Nutsedge	1.66	0.98	4.09	7.25	0.73±2.00	42.7	41.4	1.00	1.00

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Table (2): LC₅₀, LC₉₀, Slope, toxicity index and relative potency values for hexane weed extracts at different exposure periods against the adult stage of *Monacha obstructa*. (as baits)

Weed extracts	LC ₅₀ g/ml	95% confidence limits		LC ₉₀ g/ml	SE ±Slope	Toxicity index at		Relative potency at	
		Lower	Upper			LC ₅₀	LC ₉₀	LC ₅₀	LC ₉₀
hours 24 After									
Primpernel	2.05	1.12	14.9	15.0	0.57±1.48	100	100	1.14	1.19
Nightshade	2.34	1.37	12.8	17.9	0.48±1.45	87.6	83.7	1.00	1.00
Nutsedge	-	-	-	-	-	-	-	-	-
hours 48 After									
Primpernel	1.64	1.07	3.25	6.02	0.76±2.27	100	100	4.88	10.8
Nightshade	2.08	1.21	10.8	18.9	0.44±1.34	78.8	31.9	3.85	3.44
Nutsedge	8.01	-	-	65.1	1.29±1.40	20.5	9.25	1.00	1.00
hours 96A fter									
Primpernel	1.22	0.79	2.01	5.20	0.57±2.04	100	100	4.14	7.92
Nightshade	1.50	0.95	2.80	6.00	0.69±2.12	81.3	86.7	3.36	6.87
Nutsedge	5.05	-	-	41.0	0.88±1.41	24.2	12.6	1.00	1.00

Table (3): Relative susceptibility of the adult stage of the land snail, *Monacha obstructa* to chloroform and hexane extracts of given weeds (contact poisons)

Weed extracts	LC ₅₀ g/ml	95% confidence limits		LC ₉₀ g/ml	SE ±Slope	Toxicity index at		Relative potency at	
		Lower	Upper			LC ₅₀	LC ₉₀	LC ₅₀	LC ₉₀
Chloroform extract									
Nightshade	0.93	0.63	1.20	2.17	0.85±3.48	100	100	1.33	3.00
Primpernel	0.99	0.64	1.38	3.11	0.65±2.57	94.0	69.8	1.25	2.04
Nutsedge	1.24	0.75	2.26	6.34	0.53±1.80	75.0	34.2	1.00	1.00
Hexane extract									
Primpernel	1.13	0.69	30.5	4.86	0.90±2.02	100	100	1.74	1.62
Nightshade	1.63	1.17	2.76	5.21	0.76±2.54	69.3	93.2	1.21	1.51
Nutsedge	1.97	1.34	5.17	7.86	0.72±2.13	57.4	61.8	1.00	1.00

Table (4): Relative susceptibility of the adult stage of the land snail, *Monacha obstructa* to chloroform and hexane extracts of given weeds (antifeedants)

Weed extracts	SC ₅₀ g/ml	95% confidence limits		Fold	SC ₉₀ g/ml	95% confidence limits		Fold	SE ±Slope
		Lower	Upper			Lower	Upper		
Chloroform extract									
Primpernel	0.22	0.18	0.26	12.6	0.93	0.76	1.21	23.0	0.19±2.04
Nightshade	2.78	2.07	4.32	1.00	21.3	11.1	60.9	1.00	0.18±1.45
Nutsedge	-	-	-	-	-	-	-	-	-
Hexane extract									
Primpernel	0.78	0.63	0.94	5.33	3.25	2.73	4.02	9.17	0.14±1.42
Nightshade	1.15	1.02	1.28	3.62	6.24	4.33	10.6	4.78	0.21±2.85
Nutsedge	4.16	3.17	6.15	1.00	29.8	16.4	76.8	1.00	0.17±1.50

• **Identification of chemical components in the chloroform extract of primpernel by GC/MS analysis**

Fifteen compounds were identified by GC/MS, Gas chromatography analysis, in the chloroform extract of primpernel as shown in table (5) and figs. (1,2 and 3), Palmitic acid methyl ester as fatty acid (10) was the dominant compound amounting to 25.8% followed by aromadendrene as sesquiterpene (3) representing 15.2% and 2-tertbutyl - 4 - isopropyl - 5- methylphenol as phenolic compound (5) forming 13.9%. Besides, the presence of two steroids; ethyl – iso allocholate (14) and 1.3 – hydroxyspirost – 8 – en – 11 – one (13) were detected in small amounts with 1.87 and 1.05% respectively. Similarly, among the identified compounds as dicarboxylic acids were malonamic acid (1) and dibutyl phthalate (6) with 4.19 and 3.29% of the total compounds and only one ketone compound; 10 (a) methyl decahydrobenzo cycloocten – 2 (1H) – one was found in peak area 2.66% (9). Finally, the salt of carbamic acid (ammonium carbamate) with 5% area (2) and the ester of phosphoric acid (tributyl phosphate) with 4.98% area (7) were also detected. This study reveals that chemical compounds present in the chloroform extract of primpernel has potent source as a molluscicide of plant origin and may be useful to control the land snail *M. obstructa*.

Table (5): Compounds Identified in the chloroform extract of primpernel by GC/MS analysis

No.	Compounds	M. W.	R. T. (min.)	% Area	Chemicals formula	Chemical group
1	Malonamic acid	103	9.57	4.19	C ₃ H ₅ NO ₃	Dicarboxylic acid
2	Carbamic acid ammonium salt	61	9.93	5.00	CH ₃ NO ₂	Carbamate
3	Aromadendrene	204	23.99	15.2	C ₁₅ H ₂₄	Terpene
4	Germacrene -D	204	25.54	2.42	C ₁₅ H ₂₄	Terpene
5	2-tertbutyl-4- isopropyl -5- methylphenol	206	26.24	13.9	C ₁₄ H ₂₂ O	Phenol
6	Dibutyl phthalate	278	29.18	3.29	C ₁₆ H ₂₂ O ₄	Dicarboxylic acid
7	Phosphoric acid tributyl ester	266	29.56	4.98	C ₁₂ H ₂₇ O ₄ P	Phosphate
8	3-{1- hydroxy-2-(methyl amino)ethyl} phenol	167	32.55	2.13	C ₉ H ₁₃ NO ₂	Phenol
9	10 (a) methyl decahydrobenzo cycloocten-2 (1H)-one	194	33.46	2.66	C ₁₃ H ₂₂ O	Ketone
10	Palmitic acid methyl ester	270	35.20	25.8	C ₁₇ H ₃₄ O ₂	Fatty acid
11	2, 5- difluoro-4-{1-hydroxy-2-(methyl amino) ethyl}-1, 2- benzenediol	219	35.97	7.41	C ₉ H ₁₁ F ₂ NO ₃	Phenol
12	Tetra cyclopropane butanoic acid methyl ester	374	38.98	2.75	C ₂₅ H ₄₂ O ₂	Fatty acid
13	1, 3-hydroxy spirost-8-en-11-one	428	39.28	1.05	C ₂₇ H ₄₀ O ₄	Steroid
14	Ethyl – iso allocholate	436	39.62	1.87	C ₂₆ H ₄₄ O ₅	Steroid
15	9, 12, 15-octa decatrienoic acid butyl ester	334	39.74	7.35	C ₂₂ H ₃₈ O ₂	Fatty acid
	Total	-	-	100	-	-

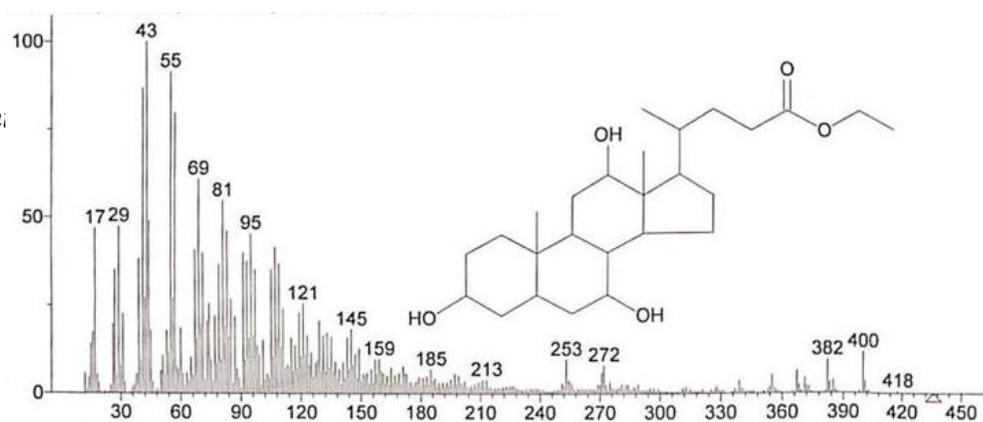


Fig. (1): Mass Spectrum and Structure of Ethyl iso-allocholate

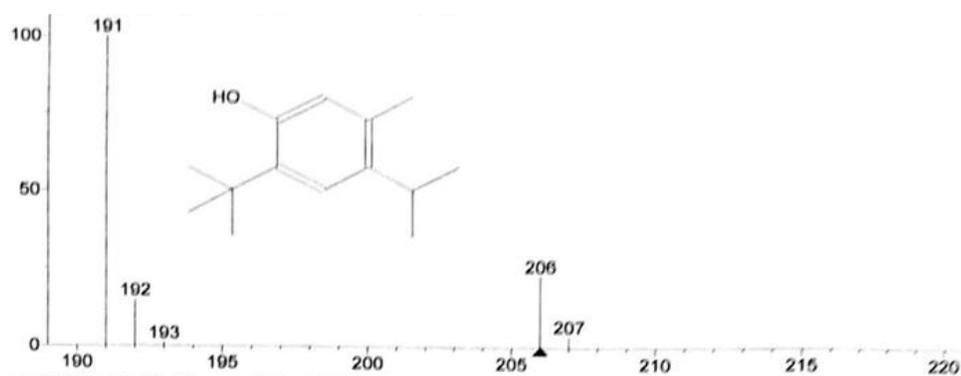


Fig. (2): Mass Spectrum and Structure of 2-tertbutyl-4-isopropyl-5-methylphenol

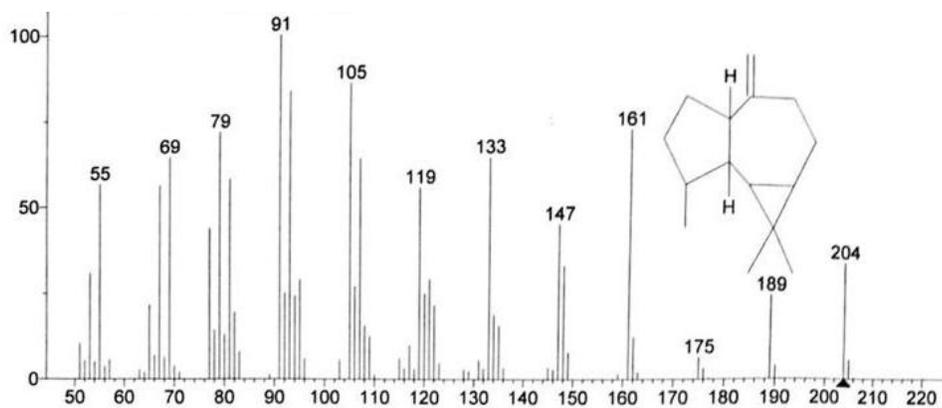


Fig. (3): Mass Spectrum and Structure of Aromadendrene

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The comparison between the total percentage of mortality of the different treatments gives a clear vision of the molluscicidal and bioactivity of the weeds extracts and their effect of the land snails *M. obstructa*.

These results are in accordance to the findings of Speiser *et al.* (1992) they found that pyrrolizidine alkaloids and sesquiterpenes extracted from the leaves of *Adenostyles alliariae* acted as feeding deterrent to young snails. Also, Hussein *et al.* (1994) reported that the shrub of *Calotropis procera* is highly toxic to the land snail, *Theba pisana*. The active ingredient responsible for the molluscicidal activity was found to be uscharin. Moreover, Dodds and Hendrson (2002) whom indentified eleven antifeedant chemicals from plants including phenols, alcohols, ketones, aldehydes and steroids, suggesting that slug or snail feeding may be controlled by these chemicals. In this respect, Ghaly *et al.* (2016) who identified sixty seven components by GC/MS in *Juniperus communis* oil with homogeneraniol (36.9%) as the major constituent while sixty components were identified in *J. horizontalis* oil with the main component, bronylacetate representing 41.2%. Two *Juniperus* species appeared molluscicidal activities.

The characteristic compounds could be useful for the terrestrial gastropods management that are effective and safe for the environment as well. More studies are needed to be tested for bioassay and the activity of the each identified compound against land snail.

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- التأثيرات الإبادية والمانعة للتغذية من بعض مستخلصات الحشائش على قوقع البرسيم الزجاجي إبراهيم حامد حسين على¹ - شيماء أحمد محمد السيد¹ - رهام فتحي على محمد² - سلوى محمد عبدالحليم¹
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- تم في هذا البحث دراسة تأثير سمية ثلاثة مستخلصات من نباتات الحشائش وهي صابون الغيط - عنب الديب - السعدة على الطور الكامل لقوقع البرسيم الزجاجي وقد تم التقييم الحيوي لهذه المستخلصات باستخدام ثلاثة أنواع من الإختبارات المعملية وهي الطعم السام - الملامسة - مضادات أو مانعات التغذية - وقد تم الإستخلاص بمذيبين مختلفين في القطبية هما الكلوروفورم والهكسان. وقد أشارت النتائج المتحصل عليها إلى النقاط التالية:-
- عندما تم إستخدام المستخلصات المختبرة كطعوم سامة على فترات تعريض مختلفة 24، 48، 96 ساعة كان مستخلص صابون الغيط الأكثر سمية على أساس قيمة LC_{50} يليه مستخلص عنب الديب في حين كان مستخلص السعدة الأقل سمية - فيما يتعلق بتأثير مستخلصات الحشائش المختبرة على قوقع البرسيم الزجاجي بإستخدام إختبار الملامسة وجد أن مستخلص الكلوروفورم لعنب الديب ومستخلص الهكسان لصابون الغيط الأكثر فاعلية في حين كان مستخلص السعدة الأقل فاعلية. من ناحية أخرى أظهر مستخلص صابون الغيط سواء في الكلوروفورم او الهكسان التأثير الأعلى كمانع للتغذية على أساس قيمة SC_{50} يليه مستخلص عنب الديب وكان مستخلص السعدة في الهكسان الأقل تأثيراً بينما نفس المستخلص في الكلوروفورم لم يعطي أى تأثير مانع لتغذية القوقع محل الدراسة.
 - كذلك تم تعريف خمسة عشر مركب كيميائي بواسطة جهاز GC/MS في مستخلص الكلوروفورم الخام لصابون الغيط كأفضل مستخلصات الحشائش المختبرة من حيث تأثيرها السام والمانع للتغذية - وقد أظهرت النتائج أن إستر الميثايل لحمض البالمتيك (حامض دهني) هو المركب السائد بنسبة 25,8% يليه الأروماديندين (تربين أحادي) بنسبة 15,2% ثم 2- بيوتانيل-4- أيزو بروبييل - 5- ميثايل فينول (مركب فينولي) بنسبة 13,9% - وقد إشتمل التعريف على إثنان من المركبات التي تتبع الأحماض ثنائية الكربوكسيل ومركبان من مجموعة الاستيرويد ومركب كيتوني واحد فقط.
 - بجانب وجود مركبات كيميائية أخرى مثل ملح حامض الكرباميك (كربامات الأمونيوم) وأستر حامض الفوسفوريك (فوسفات تراي البيوتانيل) وأخيراً قد تعزو التأثيرات السامة أو المانعة للتغذية لمستخلص صابون الغيط في الكلوروفورم إلى وجود هذه المركبات الفعالة التي تتبع مجاميع كيميائية مختلفة تشمل فينولات وكربامات وتربينات أحادية و كيتونات وأحماض ثنائية الكربوكسيل وأسترات لأحماض دهنية وفوسفورية.