

Research Article

Chemical Profiling and Insecticidal Activity of *Artemisia herba-alba* Essential Oil Against Papaya Mealybug *Paracoccus marginatus* (Hemiptera: Pseudococcidae)

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Abstract: The current study aimed to investigate the chemical composition and insecticidal activity of *Artemisia herba-alba* essential oil obtained by hydrodistillation and solvent extraction against papaya mealybug *Paracoccus marginatus*. The essential oil was analysed using gas chromatography-mass spectrometry and the insecticidal activity was recorded for different times and concentrations under laboratory conditions. According to GC-MS results, the chemical components detected of essential oil isolated by hydrodistillation and solvent extraction were 52 and 62 compounds of both extraction methods, the main components of essential oil of hydrodistillation method were eucalyptol (32.81%), α -pinene (28.62%) and linalol (10.18%) while the major components of essential oil isolated by solvent extract were pentadecanoic acid (16.69%) and dihomogamma-linolenic acid (15.2%). The essential oil of hydrodistillation method exhibited more insecticidal activity than solvent extract method against *P. marginatus*, 1000 mg/L of essential oil isolated by hydrodistillation achieved 100% mortality of papaya mealybugs after 120, 168 h of treatment with LC₅₀ values which was 65.362 mg/L after 168 h from treatment and LT₅₀ value which was 22.386 h at 1000 mg/L. The findings reveal and propose that the essential oil of *A. herba-alba* is a rich by bioactive components and it is a prospective source of botanical insecticides.

Keywords: *Artemisia herba-alba*, essential oil, eucalyptol, extraction method, GC-MS., papaya mealybug

INTRODUCTION

The papaya mealybug *P. marginatus* is a sap-sucking insect with the waxy body and considered as a devastating pest on various economically important crops. It is widely distributed on tropical and subtropical vegetables, fruits and ornamental plants. More than 55 genera are belonging to 22 plant families are the host plants for this harmful insect pests (Muniappan *et al.*, 2008; Sakthivel, 2013). The damaging results of this pest could be direct in case of sap removal and injection of toxins or indirect by honeydew contamination associated with sooty mould. It decreases the photosynthesis process which results in leaf yellowing, defoliation, retard plant growth and eventually death of plant (Tanwar *et al.*, 2010).

Several management tools have been previously applied, but still, it is one of the most challenging and difficult pests to control. Their thick waxy body cover which does not only create delinquency in penetration of pesticide, but also protect these from natural enemies. However, the chemical control remains the

first choice for papaya mealybug management, but since the exposure on the risk of chemical pest control methods to the public health; there has been an on-going attempt to reduce the use of harmful pesticides. These issues have led to the development of more targeted compounds that exhibit fewer side effects (Kocur-Bera and Dudzińska, 2014; Rasheed *et al.*, 2014). Plant materials that possess insecticidal properties are one of the most critical, biodegradable and inexpensive methods to insect control as they do not have residues toxic to the environment and humans (Ebadollahi, 2013).

Artemisia spp. is one of the vital genus belongs to the large family of Asteraceae which comprises more than 300 species, some species are aromatic plants used as food and remedy meanwhile the others are toxic. *Artemisia spp.* are grown widely in Europe, America, Asia and Australia and also can be found in the northern hemisphere (Mohsen and Ali, 2008; Tilaoui *et al.*, 2011). *A. herba-alba* also was known as desert wormwood and Al-Shih in Arabic, is aromatic herb spreads in many arid and semi-arid regions of the

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Mediterranean basin and stretches into the northwestern Himalayas. This plant is rich in essential oil and active components in comparison to other species of *Artemisia* genus. Since ancient times, this plant has been utilised by many cultures in folk medicine for therapy, intestinal disturbances, diabetes, muscle weakness, colds, coughing and an antidiabetic agent (Mohamed *et al.*, 2010; Paolini *et al.*, 2010).

Many studies have accentuated the insecticidal effects of *A. herba-alba* essential oil and extracts involving growth retardation as well as antifeedant and larvicidal effects against different types of insects. Soliman (2007) reported that *A. herba-alba* essential oils exhibited high toxicity on *Aphis gossypii*, *Thrips tabaci* and *Bemisia tabaci* under laboratory conditions. Sharifian *et al.* (2012) showed the insecticidal and biological activity of *A. herba alba* essential oil against *Tribolium castaneum*, *Callosobruchus maculatus* and *Rhyzopertha dominica*. The essential oil of *A. herba-alba* was found to have antifeedant and repellent activities against two types of stored cereal pests such as *T. castaneum* and *Trogoderma granarium* (Badreddine and Baouindi, 2016).

Despite the vital role played by essential oil or plant extract as insecticides preventing plants from being infested by insects is commonly accepted and *A. herba alba* as a vigorous plant able to manage many types of insects due to its insecticidal or repellent properties are considered. No such study has been previously carried out to record the insecticidal activity of this plant in regard to control papaya mealybug *P. marginatus*. Therefore, the present study was assigned to inspect the chemical composition of the essential oils isolated from the aerial part of *A. herba-alba* by hydrodistillation and solvent extraction. Further, it estimated its efficacy-toxicity on the female adults of *P. marginatus* in the laboratory condition.

MATERIALS AND METHODS

Insect rearing: Papaya mealybug rearing was carried out using pumpkin fruits (*Cucurbita moschata* Duchesne) method. Papaya leaves (*Carica papaya* L) infested with *P. marginatus* were collected from the field No. 2 of Universiti Putra Malaysia. These infested leaves were brought into the laboratory and further reared on medium-sized pumpkin fruits. The pumpkin fruit was washed with tap water and dried well before to release insects on it. The nymphs and egg sacs of *P. marginatus* were transferred from infested Papaya leaves to pumpkin fruits by camel hair brush. Each pumpkin was placed in a plastic container (10×20×10 cm) and kept under laboratory conditions at 27±2°C and 65±5% RH with a photoperiod of 12:12 (L: D). (Saengyot and Burikam, 2012).

Plant material: The aerial parts (leaves and stems) of *A. herba-alba* were imported from local markets of Iraq. The sample plants were cleaned of dirt and dried for 7-10 days under the shade at room temperature. The dried plants were crushed into powder using a commercial electrical mill and kept in plastic containers in a dry place until further use.

Essential oil extract: The essential oil extract was carried out by following protocols

Hydrodistillation: The procedure was followed as previously mentioned by Al-Shuneigat *et al.* (2015). About 300 g of plant sample powder was subjected to a Clavenger hydrodistillation device for 6 hours to obtain essential oils. The yield of essential oil was stored at 4°C for further use.

Solvent extraction: About 100 g of sample plant powder was soaked and shaken with 500 mL of n-hexane in a conical flask for 24 h at room temperature using an electric shaker device (Orbital shaker; Model-719). The extract was collected and separated through Whatman No. 1 filter paper, then it was purified by 0.45 µm membrane filter (Thermo Scientific Titan 3). The crude extract was placed at room temperature to allow the solvent to evaporate. The extraction yield was kept at 4°C until further use (Khosravi *et al.*, 2011).

Gas Chromatography-Massspectrometry (GC-MS) analysis: The GC-MS analysis of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction was carried out using GC-MS-QP2010 Shimadzu system including a gas chromatograph. It was interfaced to mass spectrometer instrument with the Nist library, employing the following conditions: column ZB-5MS 30 m×0.25 mm ID×0.25 µm film thickness capillary column composed of 5% (phenylmethyl polysiloxane), 0.2 µL of the sample was injected using the split mode. The carrier gas used was helium (99.99%) with a flow rate of 2.00 mL/min. The injector temperature was 300°C, ion source temperature was 240°C and the oven temperature was programmed from 40°C (isothermal for 3 min) with a rising temperature of 20°C/min to 325°C. Mass spectra were taken at 70eV, a scan interval of 0.5 sec and fragments from 40 to 440Da with total GC running time of 22 min. The individual components were recognised by comparing of both mass spectra and their GC retention data; other identifications were known by comparison of mass spectra with those in the data system libraries. The relative number of individual components was presented as a percentage by regularisation of the peak area (Martínez-Díaz *et al.*, 2015).

Laboratory bioassay: The leaf spraying method was followed to evaluate the toxicity of *A. herba-alba*

essential oil obtained by hydrodistillation and solvent extraction against *P. marginatus*. Seven different concentrations of essential oil emulsions (0, 50, 100, 200, 300, 500, 1000 mg/L) were prepared by diluting with distilled water which contained 0.5% of Triton X-100 surfactant. Papaya leaves were collected from the field and after washing and drying, the leaves were sprayed in different concentrations using a small sprayer and left to dry for one hour at room temperature. Later, these leaves were put on moisture filter paper (Whatman No. 1) inside Petri-dishes. The 20 female adults were released into each petri-dish using camel hair brush. The papaya leaves of all treatments were renewed at every 24 h. The experiment was conducted in four replicates each replicate had one Petri-dish (4.5 cm). Mortality was observed after 24, 48, 72, 120, 168 h after spraying. The adults were considered dead if they did not respond when touched by a thin camel hair brush. The bioassay was conducted at a temperature of $27\pm 2^{\circ}\text{C}$, $65\pm 5\%$ (RH) relative humidity and 14L: 10D photoperiod (Soliman, 2007; Fatima *et al.*, 2016).

Data analysis: The experiments were conducted in Complete Randomized Design (CRD). All collected data were analysed using two-way Analysis of Variance (ANOVA). Duncan test at 0.05 probabilities was used to separate the means with significant differences. The mortality was measured in percentage and corrected using Abbott's formula. The analysis was done using Statistical Analysis Software version 9.3 (SAS Institute, 2011). The Lethal Concentration (LC_{50}) and Lethal Time (LT_{50}) values were obtained from Probit regressions using the Polo-PC program.

RESULTS

The GC-MS analysis of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction afforded different composites which were 52 and 62 compounds which corresponded 99.97 and 100% of essential oil isolated by hydrodistillation and solvent extraction compounds presented respectively. The chemical profiles of essential oil were divided into different chemical groups depending on the type of terpene components as showed in Table 1. The essential oil isolated by hydrodistillation consisted of 11 monoterpenes (37.03%), 23 oxygenated monoterpenes (57.71%), three sesquiterpenes (0.33%), one oxygenated sesquiterpene (0.2%) and 14 others hydrocarbon compounds (4.70%). However, the main components were eucalyptol (32.81%) with retention time of 8.607 min, α pinene (28.62%) with retention time of 6.726 min, linalol (10.18%) with retention time of 9.764 min, d-Limonene (6.04%) in retention time 8.551 min and α .terpineol (6.02%) with retention time of 11.319 min. While, the mean group compounds of *A. herba-alba* essential oil isolated by solvent extraction included 13 oxygenated monoterpenes (9.69%), seven oxygenated diterpenes (19.75%), two sesquiterpenes (0.73%), eight oxygenated sesquiterpenes (18.82%), one triterpene (0.64%), six oxygenated triterpene (3.16%) and 23 others hydrocarbon compounds (47.21%). However, the major compounds were dihomogamma-linolenic acid (15.20%) with retention time of 13.812 min, pentadecanoic acid (13.91%) with retention time of 12.944 min, pentadecanoic acid (2.78%) with retention time of 13.901 min and phytol (2.21%) with retention time 12.302 min.

Table 1: Percentage composition of *A. herba-alba* essential oil obtained by hydrodistillation and solvent extraction

Compound	Extraction method			
	Hydrodistillation		Solvent	
Monoterpene	RT(min.)	Amount (v/v)%	RT(min.)	Amount (v/v)%
α . Phellandrene	6.592	0.07		
α .Pinene	6.726	28.62		
Camphene	7.051	0.14		
β . Pinene	7.592	0.14		
β . -Myrcene	7.829	0.5		
α .Terpinen	8.326	0.06		
β . -Cymene	8.471	0.56		
D-Limonene	8.551	6.04		
Ocimene	8.854	0.34		
α .Terpinolen	9.531	0.36		
γ .Terpinen	9.065	0.2		
Subtotal%		37.03		
Oxygenated Monoterpene				
Eucalyptol	8.607	32.81	6.829	0.78
linalool oxide	9.286	0.32		
Linalol	9.764	10.18	8.103	0.16
Pinocarveol	10.472	0.22		
Alcanfor	10.581	0.26	7.902	0.17
Linderol	10.969	0.3		
(-)-4-Terpineol	11.096	0.31		

Table 1: Continue

α . Terpineol	11.319	6.02	8.268	0.46
cis-Carveol	11.673	0.2		
Nerol	11.733	0.16		
Pulegone	11.975	0.21	8.616	0.99
Bergamiol	12.085	1.27		
Piperitone	12.208	1.09	8.737	0.55
Carvacrol	12.683	0.06	9.014	0.47
Thymol	12.797	0.29		
Verbenone	13.038	0.05		
Methyl geranate	13.108	0.11		
Myrtenyl acetate	13.167	1.92		
DHS activator	13.379	0.15		
α . Terpineol acetate	13.485	1.13		
Ethyl cinnamate	15.104	0.2		
Durohydroquinone	15.680	0.36		
Sabinol	17.164	0.09		
2,3a-Dimethylhexahydrobenzofuran-7a-ol			8.457	0.64
6-Oxocineole			9.074	0.66
Cineron			9.365	0.94
γ .Thujaplicin			10.425	0.55
Mint Lactone			10.668	2.82
Isomenthol			13.675	0.5
Subtotal%		57.71		9.69
Sesquiterpene				
Caryophyllene	14.525	0.11	9.951	0.16
α .Caryophyllene	14.993	0.17		
Eudesma-4(14),11-diene	15.423	0.05	19.043	0.57
Subtotal%		0.33		0.73
Oxygenated Sesquiterpene				
Dactylol			8.975	0.36
Spathulenol	16.501	0.2	10.959	0.32
Caryophyllene oxide			11.008	0.29
Pentadecanoic acid			12.944	13.91
Pentadecanoic acid			13.42	0.25
Pentadecanoic acid			13.901	2.78
AC1NSIAW			17.583	0.29
AC1LD5V1			17.947	0.62
Subtotal%		0.2		18.82
Oxygenated Diterpene				
Andrographolide			11.498	0.15
phytol			12.303	2.21
(2E)-3,7,11,15-Tetramethyl-2-hexadecen-1-ol			12.431	0.6
(2E)-3,7,11,15-Tetramethyl-2-hexadecen-1-ol			12.531	0.8
Manool Oxide			13.287	0.22
Dihomo- γ .linolenic acid			13.812	15.2
Eicosanoic acid			14.786	0.57
Subtotal%				19.75
Triterpene				
A-Neooleana-3(5),12-diene			16.954	0.64
Subtotal%				0.64
Oxygenated Triterpene				
Epiputranjivol			14.698	0.35
Olean-12-en-3-one #			17.7	0.66
3-epicycloeucalenol			17.746	1.12
(E)-24-Propylidenecholesterol			18.675	0.23
β . Amyrenol			18.905	0.23
Urs-12-en-28-al			20.676	0.57
Subtotal%				3.16
Others compounds		4.7		47.21
Total%		99.97		100

A significant difference ($p < 0.05$) in mortality of adult papaya mealybug at different time after treatment, type of extraction and concentration was observed. The mortality results further showed the different levels of insecticidal toxicity on insect (Table 2). The mortality

percentage increased significantly with increased treatment concentration and time after treatment. The highest (100%) mortality percent of adults with the treatment of essential oil at a maximum concentration of 500 mg/L and 1000 mg/L after 120 and 168 h

Table 2: The mortality of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction against papaya mealybug, *P. marginatus*

Concentration (mg/L)	Time after treatment (h.)				
	24	48	72	120	168
	Mean±S.E	Mean±S.E	Mean±S.E	Mean±S.E	Mean±S.E
Solvent extraction					
50	7.04±0.51	11.16±0.11	15.00±0.00	29.91±0.14	33.72±0.05
100	8.61±0.18	13.67±0.11	22.44±0.09	33.72±0.05	41.23±0.04
200	13.67±0.11	16.19±0.09	25.00±0.00	38.72±0.05	50.00±0.00
300	17.42±0.11	18.69±0.09	27.45±0.08	45.00±0.00	61.23±0.03
500	21.20±0.07	25.00±0.00	36.14± 0.16	53.73±0.03	67.44±0.10
1000	45.00±0.00	50.00±0.00	56.23±0.03	68.73±0.03	87.36±0.22
Hydrodistillation extraction					
50	17.28±0.27	25.00±0.00	37.46± 0.06	55.00±0.00	69.87±0.19
100	21.20±0.07	33.72±0.05	42.39±0.15	62.48±0.04	78.74±0.03
200	23.70±0.07	38.72±0.05	66.19±0.10	74.92±0.12	86.24±0.02
300	28.71±0.06	42.47±0.05	77.41±0.14	82.35±0.23	93.74±0.02
500	37.46±0.06	61.23±0.03	83.70±0.08	91.24±0.02	100.00±0.00
1000	60.00±0.00	72.48±0.03	94.97± 0.05	100.00±0.00	100.00±0.00

Table 3: LC₅₀ (mg/L) values of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction against papaya mealybug, *P. marginatus*

Time (hours)	LC ₅₀ (95% C.L.)*		Slope±S.E.	X ²	Heterogeneity	df.
Solvent extraction						
24	2156.89	1256.81 to 5832.29	1.05±0.17	7.52	0.34	22
48	1243.40	828.86 to 2443.76	1.09±0.16	5.20	0.23	22
72	831.07	550.75 to 1693.67	0.84±0.14	2.67	0.12	22
120	439.78	330.35 to 648.32	0.98±0.14	3.76	0.17	22
168	154.11	115.18 to 196.76	1.11±0.14	8.31	0.38	22
Hydrodistillation extraction						
24	740.80	537.47 to 1204.66	1.07±0.15	6.57	0.29	22
48	366.05	278.41 to 516.82	0.99±0.14	7.56	0.34	22
72	192.82	145.20 to 249.92	1.03±0.14	2.96	0.13	22
120	110.20	76.69 to 144.14	1.08±0.14	6.52	0.30	22
168	65.36	46.74 to 83.29	1.57±0.18	13.64	0.62	22

*C.L. (confidence limit) which has been calculated with 95% confidence

Table 4: LT₅₀ (hour) values of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction against papaya mealybug, *P. marginatus*

Concentration (mg/L)	LT ₅₀ (95% C.L.)*		Slope±S.E.	X ²	Heterogeneity	df.
Solvent extraction						
50	342.78	218.09 to 907.57	1.34±0.27	4.98	0.28	18
100	243.71	172.24 to 473.32	1.42±0.25	1.83	0.10	18
200	196.32	143.35 to 349.79	1.33±0.24	3.32	0.18	18
300	138.78	110.72 to 195.80	1.53±0.23	6.825	0.38	18
500	101.96	83.90 to 131.39	1.54±0.23	4.76	0.26	18
1000	39.74	27.12 to 50.89	1.27±0.22	11.31	0.63	18
Hydrodistillation extraction						
50	98.36	82.90 to 121.34	1.77±0.23	5.22	0.29	18
100	75.54	64.04 to 89.72	1.83±0.23	4.56	0.25	18
200	54.55	46.39 to 62.88	2.17±0.24	5.29	0.29	18
300	44.74	38.06 to 51.20	2.47±0.25	13.07	0.73	18
500	33.62	28.11 to 38.67	2.79±0.29	6.61	0.37	18
1000	22.38	17.12 to 26.81	2.93±0.38	12.36	0.69	18

*C.L. (confidence limit) which has been calculated with 95% confidence

was observed by hydrodistillation extraction. The lowest (7.04% and 8.61%) mortality percent of adults with the treatment of essential oil at a minimum concentration of 50 and 100 mg/L after 24 h was observed by solvent extraction. These results were later reflected in the LC₅₀ and LT₅₀ values which represented the toxicity levels of *A. herba-alba* essential oil isolated by both hydrodistillation and solvent extraction.

The results of log-probit analysis regarding LC₅₀ and LT₅₀ values of *A. herba-alba* essential oil showed in Table 3 and 4. According to these results, the lethal

concentration (LC₅₀) values progressively decreased with increasing the time after treatment and the lethal time (LT₅₀) reduced with raising the concentration at 95% confidence level. The essential oil of *A. herba-alba* obtained by hydrodistillation was more toxic as an insecticide against papaya mealybugs with LC₅₀ (95% C.L.) values of 65.36 mg/L (46.74 to 83.29 mg/L) and 110.20 mg/L (76.69 to 144.14 mg/L) after 168 and 120 h, respectively. Whereas the essential oil obtained by solvent extraction was recorded lowest toxic against adults papaya mealybugs with LC₅₀ values of 2156.89

mg/L (1256.81 to 5832.29 mg/L) after 24 h, followed by LC₅₀ values of 1243.40 mg/L (828.863 to 2443.76 mg/L) and 831.07 mg/L (550.75 to 1693.67 mg/L) at 95% confidence level after 48 and 72 h, respectively. Also, the better LT₅₀ (95% C.L.) values of 22.38 h (17.12 to 26.81 h) was observed of essential oil isolated by hydrodistillation at a concentration of 1000 mg/L. However, the highest LT₅₀ (95% C.L.) values of 342.78 h (218.09 to 907.57 h) of 50 mg/L concentration followed by LT₅₀ (95% C.L.) values 243.717 h (172.24 to 473.32 h) and 196.32 h (143.35 to 349.79 h) of 100 and 200 mg/L concentration was respectively observed of essential oil isolated by solvent extraction.

DISCUSSION

The major oil components observed in the present study was oxygenated monoterpenes and monoterpenes with eucalyptol, α -Pinene and linalool in the essential oil of *A. herba-alba* isolated by hydrodistillation. However, the main group compounds in the essential oil of *A. herba-alba* isolated by solvent extraction was oxygenated sesquiterpene and oxygenated diterpene with pentadecanoic acid and dihomogamma-linolenic acid. The oxygenated monoterpenes and monoterpenes were also previously reported as the main terpenes group compounds of *A. herba-alba* essential oil in different parts of the world such as Algeria, Jordan and Iran (Bezza *et al.*, 2010; Tilaki *et al.*, 2013; Al-Shuneigat *et al.*, 2015). Similarly, eucalyptol was mentioned as the major component in the essential oil of *A. herba-alba* from Morocco, Egypt, Iran and Tunis (Soliman, 2007; Mohsen and Ali, 2009; Behtari *et al.*, 2011; Talbaoui *et al.*, 2012). The present study displayed that the α -Pinene and linalool were the major components of *A. herba-alba* essential oil; the similar compounds were also found in other studies with less quantity in *A. herba-alba* collected from various geographies (Mohsen and Ali, 2009; Bellili *et al.*, 2016; Rekkab *et al.*, 2016). Al-Shuneigat *et al.* (2015) confirmed that the essential oil isolated from *A. herba-alba* grown in different countries and different areas within the same country have a high variation in chemical compounds. The variability of *A. herba-alba* active compounds can be ascribed to various factors such as environmental factors like weather condition, water and soil type, harvest time, plant part, plant age, type of plant sample used (fresh or dried), geographical factors(location), sample collection time, genetic variability (chemotype) and method of extraction (Behtari *et al.*, 2011; Tilaoui *et al.*, 2015; El-Zaeddi *et al.*, 2016; Ouaritini *et al.*, 2016).

Similarly, the results of the present study exhibited the differences in chemical composition of *A. herba-alba* essential oil isolated by hydrodistillation and solvent extraction. The essential oil isolated by hydrodistillation comprised more amount of

monoterpene and monoterpenoid compounds as compared to the essential oil isolated by solvent extraction which included on oxygenated sesquiterpene and oxygenated diterpene compounds. These findings agree with many previous studies those confirmed that the method of extracting essential oils affected the chemical components of oil (Presti *et al.*, 2005; Azad *et al.*, 2014; Maimulyanti and Prihadi, 2016). The variations in the amount and type of the chemical compositions may be due to the effect of the temperature used in extraction; as a high temperature leads to break up the naturally occurring high molecular weight of compounds in the plant sample in order to make oil containing more volatile compounds with less molecular weight (Charles and Simon, 1990; Jaenson *et al.*, 2005). The oil obtained from the solvent extract included pigments, oxygenated aliphatic compounds and colours with high molecular weight (Sukatta *et al.*, 2009; Azad *et al.*, 2014). Presti *et al.* (2005) maintained that the decrease in volatile compounds in solvent extraction might be attributed losses of more volatile compounds during the elimination of the solvent under room temperature.

The essential oil of *A. herba-alba* showed the excellent toxicity properties against adults of papaya mealybugs. Many studies have confirmed the results of the present study which indicated that the essential oil and solvent extracts of *A. herba-alba* have an insecticidal effect on numerous species of insect (Soliman, 2007; Sharifian *et al.*, 2012; Bachrouch *et al.*, 2015; Billal *et al.*, 2015; Badreddine and Baouindi, 2016). The volatile compounds such as monoterpenes and monoterpenoids in essential oil of the aromatic plant have a toxic and killer effect against insects; this toxicity increases with the prevalence of these components in the essential oil (Rajendran and Sriranjini, 2008; Khorram *et al.*, 2011). Maciel *et al.* (2010) mentioned that terpenoid components in essential oil have repellent, antifeedant, growth inhibition, reduced reproductive capacity, impaired maturation and direct toxicity properties against insects. The monoterpenes and monoterpenoids are usually volatile and lipophilic components that can quickly penetrate into the insect cuticle and intervene with their physiological functions (Erler, 2005; Tayoub *et al.*, 2012). In general, these compounds influence the nerve cell in the nervous system and inhibit the acetylcholine in insects, the mode of action of these compounds is varied depending on the type of compound and the treated insect (Bakkali *et al.*, 2008; Tayoub *et al.*, 2012). The toxic effects of *A. herba-alba* could be attributed to principal components or all components and the possible antagonistic and synergistic influence of compound (s) in oils for instance eucalyptol and α -pinene have an insecticidal effect against *Sitophilus oryzae* (Lee *et al.*, 2004). Meanwhile, Kordali *et al.* (2006) found that the eucalyptus had more toxicity against the *S. granariesas* compared to other

components. Tripathi *et al.* (2001) reported that eucalyptol compound isolated from *Artemisia annua* had contact, fumigant and antifeedant toxic activity against *Tribolium castaneum*. Bachrouch *et al.* (2015) on the other hand, confirmed that the insecticidal activity of *Artemisia spp.* essential oil cannot be interpreted by the action of their major components only. Nevertheless, these actions are the result of a synergistic interaction between all the elements. The high toxicity of essential oil isolated by hydrodistillation possibly attributed to the significant amount of volatile compounds in the oil, which considered more toxicity and effect on the insect as compared to essential oil isolated by solvent extraction as indicated by previous studies. The results of the present study agree with the previous studies which found increased mortality and toxicity of essential oil with increased time and concentration of treatments (Sharifian *et al.*, 2012; Billal *et al.*, 2015; Badreddine and Baouindi, 2016).

CONCLUSION

In the current study, hydrodistillation and solvent extraction were selected to isolate the essential oil from aerial parts of *A. herba-alba*. The essential oils were analysed by GC-MS to determine the chemical compositions of both extraction methods and further evaluated their insecticidal activity against papaya mealybugs in the laboratory. The results showed that the chemical profile was different for each extraction method. The essential oils extracted by both extraction methods were adequate to control papaya mealybugs. The essential oil isolated by hydrodistillation was more toxic against papaya mealybugs as compared to essential oil isolated by solvent extraction. The essential oil could be considered as an alternative control of papaya mealybugs as a synthetic insecticide. Nevertheless, the future experiments will focus on fractionating the different elements and then evaluate the different compounds against various types of insects to determine the component(s) accountable for the insecticidal activities.

REFERENCES

Al-Shuneigat, J., S. Al-Sarayreh, M. Al-Qudah, I. Al-Tarawneh, Y. Al-Saireh and A. Al-Qtaitat, 2015. GC-MS analysis and antibacterial activity of the essential oil isolated from wild *Artemisia herba-alba* grown in South Jordan. *Brit. J. Med. Med. Res.*, 5(3): 297-302.

Azad, F.R., A. Shafaghat and F. Salimi, 2014. Chemical constituents, antioxidant and antimicrobial activities of essential oil and hexane extract from *Arnebia linearifolia* DC. seeds. *J. Essent. Oil Bear. Pl.*, 17(1): 87-94.

Bachrouch, O., N. Ferjani, S. Haouel and J.M.B. Jemâa, 2015. Major compounds and insecticidal activities of two Tunisian *Artemisia* essential oils toward two major coleopteran pests. *Ind. Crop. Prod.*, 65: 127-133.

Badreddine, B.S. and M. Baouindi, 2016. Effects of *Artemisia herba-alba* essential oils on survival stored cereal pests: *Tribolium castaneum* (Herbst) (Coleoptera, Tenebrionidae) and *Trogoderma granarium* (Everst) (Coleoptera, Dermestidae). *J. Coastal Life Med.*, 4(5): 390-394.

Bakkali, F., S. Averbeck, D. Averbeck and M. Idaomar, 2008. Biological effects of essential oils-a review. *Food Chem. Toxicol.*, 46(2): 446-475.

Behtari, B., G.A.D. Tilaki, F. Gholami and R.B. Balkhkanlou, 2011. Comparison of the essential oil constituents of *Artemisia herba-alba* Asso in the vegetative and flowering stages. *Agr. Sci. Digest*, 31(2): 100-105.

Bellili, S., W. Dhifi, A.B.K. Al-Garni, G. Flamini and W. Mnif, 2016. Essential oil composition and variability of *Artemisia herba-alba* Asso. growing in Tunisia: Comparison and chemometric investigation of different plant organs. *J. Appl. Pharm. Sci.*, 6(7): 38-42.

Bezza, L., A. Mannarino, K. Fattarsi, C. Mikail, L. Abou, F. Hadji-Minaglou and J. Kaloustian, 2010. Chemical composition of the essential oil of *Artemisia herba-alba* issued from the district of Biskra (Algeria). *Phytothérapie*, 8(5): 277-285.

Billal, N., F. Naama and A. Imane, 2015. Insecticidal activity of three plants extracts against *Myzus persicae* (Sulzer, 1776) and their phytochemical screening. *Acta Agric. Slovenica*, 105(2): 261-267.

Charles, D.J. and J.E. Simon, 1990. Comparison of extraction methods for the rapid determination of essential oil content and composition of basil. *J. Am. Soc. Hortic. Sci.*, 115(3): 458-462.

Ebadollahi, A., 2013. Plant essential oils from apiaceae family as alternatives to conventional insecticides. *Ecol. Balkanica*, 5(1): 149-172.

El-Zaeddi, H., J. Martínez-Tomé, Á. Calín-Sánchez, F. Burló and Á.A. Carbonell-Barrachina, 2016. Volatile composition of essential oils from different aromatic herbs grown in mediterranean regions of Spain. *Foods*, 5(2): 41-54.

Erler, F., 2005. Fumigant activity of six monoterpenoids from aromatic plants in Turkey against the two stored-product pests confused flour beetle, *Tribolium confusum*, and Mediterranean flour moth, *Ephesia kuehniella*. *J. Plant Dis. Protect.*, 112(6): 602-611.

Fatima, S., M. Hussain, S. Shafqat, M. Faheem Malik, Z. Abbas, N. Noureen and Noor ul Ane, 2016. Laboratory evaluation of different insecticides against hibiscus mealybug, *maconellicoccus hirsutus* (Hemiptera: Pseudococcidae). *Scientifica*, 2016: 1-7.

- Jaenson, T.G., K. Pålsson and A.K. Borg-Karlson, 2005. Evaluation of extracts and oils of tick-repellent plants from Sweden. *Med. Vet. Entomol.*, 19(4): 345-352.
- Khorram, M.S., N.T. Nasabi, S. Jafarnia and S. Khosroshahi, 2011. The toxicity of selected monoterpene hydrocarbons as single compounds and mixtures against different developmental stages of Colorado potato beetle, *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae). *J. Entomol.*, 8(5): 404-416.
- Khosravi, R., J.J. Sendi, M. Ghadamyari and E. Yezdani, 2011. Effect of sweet wormwood *Artemisia annua* crude leaf extracts on some biological and physiological characteristics of the lesser mulberry pyralid, *Glyphodes pyloalis*. *J. Insect Sci.*, 11: 156.
- Kocur-Bera, K. and M. Dudzińska, 2014. Spatial conditions of environmental risk posed by obsolete pesticides-case study of the "Green Lungs of Poland" area. *Pol. J. Environ. Stud.*, 23(3): 763-772.
- Kordali, S., I. Aslan, O. Çalmaşur and A. Cakir, 2006. Toxicity of essential oils isolated from three *Artemisia* species and some of their major components to granary weevil, *Sitophilus granarius* (L.) (Coleoptera: Curculionidae). *Ind. Crop. Prod.*, 23(2): 162-170.
- Lee, B.H., P.C. Annis, F. Tumaalii and W.S. Choi, 2004. Fumigant toxicity of essential oils from the Myrtaceae family and 1,8-cineole against 3 major stored-grain insects. *J. Stored Prod. Res.*, 40(5): 553-564.
- Maciel, M.V., S.M. Morais, C.M. Bevilaqua, R.A. Silva, R.S. Barros, R.N. Sousa, L.C. Sousa, E.S. Brito and M.A. Souza-Neto, 2010. Chemical composition of *Eucalyptus spp.* essential oils and their insecticidal effects on *Lutzomyia longipalpis*. *Vet. Parasitol.*, 167(1): 1-7.
- Maimulyanti, A. and A.R. Prihadi, 2016. Chemical composition of essential oil and hexane extract and antioxidant activity of various extracts of *Acmella uliginosa* (Sw.) Cass flowers from Indonesia. *Agr. Nat. Resour.*, 50(4): 264-269.
- Martínez-Díaz, R.A., A. Ibáñez-Escribano, J. Burillo, L.D.L. Heras, G.D. Prado, M.T. Agulló-Ortuño, L.F. Julio and A. González-Coloma, 2015. Trypanocidal, trichomonocidal and cytotoxic components of cultivated *Artemisia absinthium* Linnaeus (Asteraceae) essential oil. *Mem. Inst. Oswaldo Cruz*, 110(5): 693-699.
- Mohamed, A.E.H.H., M.A. El-Sayed, M.E. Hegazy, S.E. Helaly, A.M. Esmail and N.S. Mohamed, 2010. Chemical constituents and biological activities of *Artemisia herba-alba*. *Rec. Nat. Prod.*, 4(1): 1-25.
- Mohsen, H. and F. Ali, 2008. Study of genetic polymorphism of *Artemisia herba-alba* from Tunisia using ISSR markers. *Afr. J. Biotechnol.*, 7(1): 44-50.
- Mohsen, H. and F. Ali, 2009. Essential oil composition of *Artemisia herba-alba* from southern Tunisia. *Molecules*, 14(4): 1585-1594.
- Muniappan, R., B.M. Shepard, G.W. Watson, G.R. Carner, D. Sartiami, A. Rauf and M.D. Hammig, 2008. First report of the papaya mealybug, *Paracoccus marginatus* (Hemiptera: Pseudococcidae), in Indonesia and India. *J. Agr. Urban Entomol.*, 25(1): 37-40.
- Ouaritini, Z.B., M. Chraïbi, K.F. Benbrahim, R. Ezziouli, S. Boukhira, N. Darkaoui and H. Derwich, 2016. Phytochemical and antibacterial activity of the essential oil of *Artemisia herba-alba* from Morocco. *J. Pharm. Biol. Sci.*, 11(6): 40-45.
- Paolini, J., E.M.E. Ouariachi, A. Bouyanzer, B. Hammouti, J.M. Desjobert, J. Costa and A. Muselli, 2010. Chemical variability of *Artemisia herba-alba* Asso essential oils from East Morocco. *Chem. Pap.*, 64(5): 550-556.
- Presti, M.L., S. Ragusa, A. Trozzi, P. Dugo, F. Visinoni, A. Fazio, G. Dugo and L. Mondello, 2005. A comparison between different techniques for the isolation of rosemary essential oil. *J. Sep. Sci.*, 28(3): 273-280.
- Rajendran, S. and V. Sriranjini, 2008. Plant products as fumigants for stored-product insect control. *J. Stored Prod. Res.*, 44(2): 126-135.
- Rasheed, M., S. Bushra and M. Tariq, 2014. Use and impact of insecticides in mealybug control. *Int. J. Adv. Biol.*, 1(2): 1-11.
- Rekkab, S., I. Abaza, S. Chibani, A. Kabouche and Z. Kabouche, 2016. Chemical composition of the essential oil of aerial parts of *Artemisia herba-alba* Asso. from Oum El-Bouaghi (Algeria) and chemotaxonomic survey. *J. Mater. Environ. Sci.*, 7(12): 4383-4390.
- Saengyot, S. and I. Burikam, 2012. Bionomics of the apestly, *Spalgis epius* (Lepidoptera: Lycaenidae), predatory on the papaya mealybug, *Paracoccus marginatus* (Hemiptera: Pseudococcidae), in Thailand. *Sonklanakarin J. Sci. Technol.*, 34(1): 1-7.
- Sakthivel, N., 2013. Effectiveness of three introduced encyrtid parasitic wasps (*Acerophagus papayae*, *Anagyrus loecki* and *Pseudleptomastix mexicana*) against papaya mealybug, *Paracoccus marginatus*, infesting mulberry in Tamil Nadu. *J. Biopesticides*, 6(1): 71-76.
- SAS Institute, 2011. SAS/IML 9.3 user's guide. Sas Institute.
- Sharifian, I., S.M. Hashemi, M. Aghaei and M. Alizadeh, 2012. Insecticidal activity of essential oil of *Artemisia herba-alba* Asso against three stored product beetles. *Biharean Biologist*, 6(2): 90-93.

- Soliman, M.M.M., 2007. Phytochemical and toxicological studies of *Artemisia* L. (Compositae) essential oil against some insect pests. Arch. Phytopathol. Plant Protect., 40(2):128-138.
- Sukatta, U., P. Rugthaworn, P. Punjee, S. Chidchenchey and V. Keeratinijakal, 2009. Chemical composition and physical properties of oil from Plai (*Zingiber cassumunar* Roxb.) obtained by hydro distillation and hexane extraction. Kasetsart J. Nat. Sci., 43: 212-217.
- Talbaoui, A., N. Jamaly, M. Aneb, A.I. Idrissi, M. Bouksaim, S. Gmouh, S. Amzazi, M. El Moussaouiti, A. Benjouad and Y. Bakri, 2012. Chemical composition and antibacterial activity of essential oils from six Moroccan plants. J. Med. Plants Res., 6(31): 4593-4600.
- Tanwar, R.K., P. Jeyakumar and S. Vennila, 2010. Papaya Mealybug and its Management Strategies. National Centre for Integrated Pest Management, New Delhi, India.
- Tayoub, G., A. Odeh and I. Ghanem, 2012. Chemical composition and fumigation toxicity of *Laurus nobilis* L. and *Salvia officinalis* L. essential oils on larvae of khapra beetle (*Trogoderma granarium* Everts). Herba Pol., 58(2): 26-37.
- Tilaki, G.A.D., R.B. Balkhanloo, M.B. Rezaee and M. Amirkhani, 2013. Allelopathic effect of *Artemisia herba-alba* Asso. essential oil on seed germination of *Agropyron desertorum* and *Agropyron cristatum*. J. Med. Plants By-Prod., 2(2): 171-176.
- Tilaoui, M., H.A. Mouse, A. Jaafari and A. Zyad, 2015. Comparative phytochemical analysis of essential oils from different biological parts of *Artemisia herba alba* and their cytotoxic effect on cancer cells. PloS One, 10(7): e0131799.
- Tilaoui, M., H.A. Mouse, A. Jaafari, R. Aboufatima, A. Chait and A. Zyad, 2011. Chemical composition and antiproliferative activity of essential oil from aerial parts of a medicinal herb *Artemisia herba-alba*. Rev. Bras. Farmacogn., 21(4): 781-785.
- Tripathi, A.K., V. Prajapati, K.K. Aggarwal and S. Kumar, 2001. Toxicity, feeding deterrence, and effect of activity of 1,8-cineole from *Artemisia annua* on progeny production of *Tribolium castaneum* (Coleoptera: Tenebrionidae). J. Econ. Entomol., 94(4): 979-983.