ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

Essential Oils as Green Insecticides: GC/MS Analysis and Toxicological Studies on Cotton Mealybugs *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcidae)

Saad, L.H.E.[1]; Negm, S.El.[2]; Saleh, A.A.[2] and Abd El-Mageed, A.E.M.[1]
[1] Plant Protection Research Institute, Agricultural Research Center, Dokki, Giza, Egypt.
[2] Pesticides Department, Faculty of Agriculture. Mansoura University, Egypt.

ABSTRACT

This study was conducted to evaluate the toxic effect of three essential oils extracted from Rosemary *Salvia rosmarinus* (Lamiales: Lamiaceae), Lemongrass *Cymbopogon citratus* (Poales: Poaceae), and Camphor *Eucalyptus melliodora* (Myrtales: Myrtaceae) leaves compared to Diver® 97% E.C. under laboratory conditions on 3rd instar nymph of Cotton Mealybug (CMB) *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae). The chemical composition of the extracted essential oils was clarified. Results showed that the most remarkable toxic essential oils to the 3rd instar nymph of *P. solenopsis* after three days of treatment were *S. rosmarinus* followed by *C. citratus*. The LC_{50} values were 3102.591 and 3323.293 ppm, respectively; while, the LC_{50} values after seven days for *C. citratus* followed by *S. rosmarinus* were 680.073 and 740.591 ppm, respectively. Gas Chromatography-Mass Spectrometry was used to analyze the essential oils and identify the most active ingredients. Results revealed that the most abundant constituents of *E. melliodora* were found to be (z)-tagetenone (20.56%) and *p*-cymene (16.62%). The *S. rosmarinus* essential oil mainly consisted of 1,8-cineol (18.37%), bornyl acetate (13.02%), and norbornan-2-one (12.53%), while the major constituent of *C. citratus* essential oil was ɤ-citral (70.44%). *S. rosmarinus* and *C. citratus* essential oil showed a significant relative

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

percentage of monoterpenes (90.87% and 89.06%), respectively, indicating this component may responsible for the highly insecticidal properties against *P.solenopss*.

Keywords: Essential oils, *Phenacoccus solenopsis*, *Salvia rosmarinus*, *Cymbopogon citratus*, *Eucalyptus melliodora*, Gas Chromatography-Mass Spectrometry

INTRODUCTION

The cotton plant (*Gossypium hirsutum* L.) is listed in the top ten crops commercialized daily. Cotton is sown in more than 100 countries; it takes about 2.5 percent of the world's arable land (OECD/ FAO, 2016). As normal to crops, pests could be a major concern for cotton affecting their yields by almost 40% if not adequately managed. In Egypt, cotton is infested by several pests during its different growth stages **(Alakhdar *et al.*, 2021)**. It is one of the most important cash crops and also called silver fiber and plays a pivotal role in the economy of the country **(Abro *et al.*, 2004)**.

Cotton Mealybugs (CMB) is a major sucking pest that attacks different varieties of cotton **(Sahito *et al.*, 2009)**. In spite of large acreage, the yield of seed cotton is very low because of severe pest complexes. In addition to the direct losses that the insects can cause by sucking the phloem sap, its feeding secretions (honeydew) cause additional losses to the plants by disturbing the photosynthesis activity and inducing fungal contaminations **(Arif *et al.*, 2012)**. The honeydew also attracts ants which in turn helps in dispersion from plant to plant. Ants also protect mealybugs from predatory ladybird beetle, parasites, and other natural enemies **(Tanwar *et al.*, 2007)**. Mealybugs "hard to kill pests" commonly known as Pseudococcids are ubiquitous groups of sap-sucking plant insects. CMB is a highly polyphagous insect pest; it attacks more than 154 plant species including field crops, vegetables,

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

ornamentals, weeds, bushes, and trees **(Arif *et al.*, 2009 and Saini *et al.*, 2009)**. It causes economic damage mainly to cotton, brinjal, okra, tomato, sesame, sunflower, and China rose **(Arif *et al.*, 2009)**. The infested cotton plants become stunted, weak, and produce only a few small bolls. The leaves appear distorted, turn yellow and eventually drop off **(Culik and Gullan, 2005)**.

Many studies have determined the importance of using natural alternatives in integrated pest management programs (**Alakhdar and Ghareeb, 2021**), to minimize the overuse of traditional pesticides with emphasis on the integrated use of biological control, selective pesticides, and plant derivatives which are safe to the non-target organisms and the environment. The infestations with mealybugs on different host plants could be effectively controlled using biological control agents **(Mohyuddin *et al.*, 1997)**, plant extracts **(Dinesh *et al.*, 2003** and **Sunitha *et al.*, 2009)**, homeo chemicals **(Ahmad *et al.*, 2011)**, and synthetic insecticides **(Gross *et al.*, 2001** and **Suresh *et al.*, 2010)**.

Botanical pesticides are biodegradable **(Devlin and Zettel, 1999)** and their use in crop protection is a practical sustainable alternative nontoxic product and potentially suitable for use in integrated management programs **(Mossa, 2016)**. The use of phytochemicals (natural alternatives of plant origin) as a possible alternative to chemical insecticides might be one of such options which should be explored given their availability, potency, and low-cost effect **(Alagarmalai, 2017)**. So, the ultimate goal is to evaluate the toxic effect of three extracted leaves essential oils of Rosemary *Salvia rosmarinus*, Lemon grass *Cymbopogon citratus*, and Camphor *Eucalyptus melliodora* compared to Diver® 97% E.C under laboratory conditions on 3rd instar nymph of cotton mealybug and clarify the chemical composition of the essential oils.

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

MATERIALS AND METHODS

Tested compounds:

1-Mineral oil:

Diver oil® 97% E.C. is light mineral oil (summer oil) produced by El-Helb pesticides and chemical Company as emulsified concentrate formulation. It was used at a rate of 1.5%. Source: Plant Protection Institute, Cairo.

2-Phytochemicals (Essential oils):

Rosemary *Salvia rosmarinus* (Family: Lamiaceae).

Lemon Grass *Cymbopogon citratus* (Family: Poaceae).

Camphor Tree *Eucalyptus melliodora* (Family: Myrtaceae).

Tested species:

The Cotton Mealybug "CMB" *Phenacoccus solenopsis* (Tinsley) belongs to phylum Arthropoda, class Insecta, order Hemiptera and family Pseudococcidae.

Toxicological studies:

Culture of *Phenacoccus solenopsis* Tinsley:

A laboratory strain of the cotton mealybug *P. solenopsis* was maintained under constant conditions of (30±2 ºC, 65±5 %RH) and kept off any contamination with chemicals till the time of study to obtain a homogenous strain. *P. solenopsis* was reared on sprouted potato *Solanum tuberosum* as described by **(Rashid *et al.*, 2011)**.

Essential oils extraction technique:

Leaves (1500 gm. from each plant) were collected in January 2020 from the farm of Medicinal Plants, Pharmacognosy Department, Faculty of Pharmacy, Mansoura University. The experiment was conducted at (Plant Protection Research Institute "PPRI"), Mansoura. In laboratory-scale techniques, steam distillation is the common method used for essential oils (EOs) extraction. In this method, "Cleavenger Apparatus" was used. The type of distillation operation, Namely: Direct Steam

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

Distillation that used to treat quantities of fresh plant material described by **Masango (2005)**. The freshly aromatic plant's leaves were subjected to hydro-distillation for 8 hrs in a Clevenger-type apparatus to extract their vaporizing essential oils and stored in dark glass tubes under refrigeration (4°C) until use described by **Mostafa *et al.* (2018)**.

Gas Chromatography-Mass Spectrometer analysis of essential oils: The essential oil extracted from leaves of (*E. melliodora*, *S. rosmarinus* and *C. citratus*) were analyzed by a gas chromatograph coupled with mass spectrometer "GC/MS" in "NOWAH Scientific Center – Cairo – Egypt". Gas chromatography-mass spectrometry instrument stands with the following specifications, Instrument: a TRACE GC Ultra Gas Chromatographs (THERMO Scientific Corp., USA), coupled with a Thermo mass spectrometer detector (ISQ Single Quadrupole Mass Spectrometer). The GC-MS system was equipped with a TR-5 MS column (30 m x 0.32 mm i.d., 0.25 µm film thickness). Analyses were carried out using helium as carrier gas at a flow rate of 1.0 mL/min and a split ratio of 1:10 using the following temperature program: 60 C for 1 min; rising at 4.0 C/min to 240 C and held for 1min. The injector and detector were held at 210^{o}C. Diluted samples (1:10 hexane, v/v) of 1µL of the mixtures were always injected. Mass spectra were obtained by electron ionization (EI) at 70 eV, using a spectral range of m/z 40-450. The identification of the chemical constituents of the essential oil was de-convoluted using AMDIS software (www.amdis.net) and identified by its retention indices (relative to n-alkanes C_8-C_{22}), mass spectrum matching to (authentic standards (when available), Wiley spectral library collection, and NSIT library database as described by **(Robert, A. 1995)**.

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

Bioassay:

For determination of the median lethal concentrate (LC_{50}) values, a serial of concentrations (500, 700, 900, 1500, and 2000 ppm) and (500, 2000, 5000, 7000, and 12000 ppm) was prepared from essential oils and pesticide, respectively. Fresh cotton leaves were collected and used as a food source. These leaves were dipped for 20 sec. in each concentration then left to dry for 30 min. The leaves were dipped in distilled water only in control. Ten newly moulted 3rd instar nymphs of cotton mealybugs were transferred to each Petri dish using a fine camel hair brush, representing one replication, then confined with treated leaves in glass Petri plates. Treated leaves had been removed and fresh untreated leaves were provided for another day. Three replications were made for each concentration and the control. The dishes were covered with their lids to prevent the insect escape and maintained under constant conditions. A binocular microscope was used to distinguish dead insects from live ones. The daily inspection was carried out of all treatments (Number of dead insects) and the mortality percentages were recorded after 3 and 7 days of treatment.

-Statistical analysis:

The average mortality percentage was corrected by the following formula described by **(Schneider-Orelli's, 1947)** and **(Puntener, 1981)**.

$$\text{Corrected \% mortality} = \frac{\text{\%mortality in treatment-\%mortality in control}}{\text{100- \%mortality in the control}} * 100$$

From which the corresponding concentration probit lines (LC-p lines) were estimated in addition to determining (LC_{50} - LC_{90}) and slope values of the tested compounds were also estimated. In addition, toxicity index was measured for each tested compound using **(Sun's equation, 1950)** as follows:

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

$$\text{Toxicity Index} = \frac{LC_{50} \text{ of the most effective compound}}{LC_{50} \text{ of the tested compound}} * 100$$

RESULTS AND DISCUSSION

Three plant essential oils of three different families were extracted and examined for their insecticidal activity against 3rd instar nymph of *P. solenopsis* (Tinsley). The susceptibility of *P. solenopsis* 3rd instar nymph to the tested essential oils **Table (1)** showed that *S. rosmarinus* exhibited a high degree of efficiency as insecticide after three days of initial application followed by *C. citratus*, *E. melliodora*, and Diver oil® showing the $LC_{50's}$ of (3102.591, 3323.293, 4966.815 and 15080.433) ppm, respectively and $LC_{90's}$ of (13226.041, 17074.043, 22656.729 and 95566.056) ppm, respectively. Based on *S. rosmarinus* (100), the toxicity index being (93.359, 62.466, and 20.574) % for *C. citratus*, *E. melliodora* and Diver oil®, respectively. However, in **Table (2)** *C. citratus* proved a high degree of efficiency as insecticides after seven days of application followed by *S. rosmarinus*, Diver oil® and *E. melliodora* showing the $LC_{50's}$ of (680.073, 740.591, 1381.831, and 2483.432) ppm, respectively and $LC_{90's}$ of (4020.773, 4226.784, 8974.716 and 13385.587) ppm, respectively. Based on *C. citratus* (100), the toxicity index being (91.828, 49.215, and 27.384) % for *S. rosmarinus*, Diver oil®, and *E. melliodora*, respectively.

Li *et al.* (2021) revealed that intercropping rosemary with sweet pepper under greenhouse conditions significantly reduced the population densities of three major pest species on sweet pepper, *Frankliniella intonsa*, *Myzus persicae*, and *Bemisia tabaci*, but did not affect the population densities of released natural enemies, predatory bug *Orius sauteri*, and parasitoid *Encarsia formosa*. **Habibpour *et al.*, (2020)** showed that LC_{50} values of pure *Mentha longifolia* L. and *Mentha piperita* L. EOs (F: Lamiaceae) after 48 hours were 113.49 and 129.74 ppm, respectively on reared *P. solenopsis* 1st instar nymphs.

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

Other studies **(El-Sonbaty *et al.*, 2018)** recorded 100 and 100 % mortality at high concentrations (40µl/l) against females of *P. solenopsis*, for Thyme (F: Lamiaceae) and Lemon grass (F: Poaceae), respectively. However, Thyme essential oil recorded the highest mortality percentage with LC_{50} = 8.094 µl/l followed by Lemon grass. Results are in agreement with those obtained by (**Mostafa *et al.*, 2018)** who proved that *Thymus vulgaris* (F: Lamiaceae) exhibited a high degree of efficiency as insecticide after 24h and 72h of initial application against *P. solenopsis* adult females followed by *Syzygium aromaticum* (F: Myrtaceae). Also, **Mohamed *et al.* (2018)** found that mortality varied according to the essential oil type and the delivered dose (ppm). The most remarkable toxic essential oils after 24h and 72h of treatment were *T. vulgaris* (F: Lamiaceae) followed by *Mentha longifolia* (F: Lamiaceae) essential oil. The LC_{50} values were 29.03 and 34.32 ppm, respectively after 24h while after 72h of treatments were 15.04 and 24.93 ppm, respectively. Similar results were also documented by **(Peschiutta *et al.*, 2017)** who evaluated *Minthostachys verticillata* Griseb. (F: Lamiaceae) and *Eucalyptus globulus* Labill. (F: Myrtaceae) EOs as insecticidal products on *Planococcus ficus* Signoret under laboratory conditions. Results revealed that *M. verticillata* (LC_{50} 39.60 µL.L-1) was more toxic than *E. globulus* (LC_{50} 63.97 µL·L-1).

Prishanthini and Vinobaba (2014) obtained that *Ocimum sanctum* L. (F: Lamiaceae) was effective significantly ($p<0.05$) at lower concentrations and has the 0.6% concentration as LC_{50} against *P. solenopsis*. **Hayat *et al.* (2015)** indicated that the neem seed extracts (acetone and n-hexane extracts) caused 100 % mortality of *P. solenopsis* after 48 hrs. **Choi *et al.* (2003)** recorded at $2.3x10^{-3}$ µl/ml air, > 80% mortality with lemongrass oil against whitefly *Trialeurodes vaporariorum* Westwood. **Muhammad *et al.* (2017)** cleared that lesser concentrations of botanicals can be used to manage cotton mealybug,

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

Phenacoccus solenopsis Tinsley, and have non-toxic effects on natural enemies.

The neurotoxic mode of action was reported by observing its symptoms when insect pests were treated with essential oils or their constituents **(Kostyukovsky *et al.*, 2002** and **Priestley *et al.*, 2003)**. The competitive inhibition of Acetyl Cholinesterase Enzyme (AChE) by monoterpenes has been previously reported **(Lee *et al.*, 2001** and **Abdelgaleil *et al.*, 2009)**. For this reason, the components of the three essential oils *S. rosmarinus*, *C. citratus* and *E. melliodora* were chemically investigated using GC/MS technique represented in **Figures (3-5)** and **Table (3)**. The qualitative and quantitative compositions of the essential oils were analyzed also in **(Fig. 3-5** and **Table 3)** and the most abundant constituents of *E. melliodora* were found to be (z)-tagetenone (20.56%), *p*-cymene (16.62%) and (-)-spathulenol (14.60%). The *S. rosmarinus* essential oil mainly consisted of 1,8-cineol (18.37%), (-)-spathulenol (5.79%), and (+)-limonen (3.17%), while the major constituents of *C. citratus* essential oil were ɤ-citral (70.44%) and γ-dodecalactone (9.22%). *S. rosmarinus* and *C. citratus* essential oil **(Table 3)** showed a significant relative percentage of monoterpenes (90.87% and 89.06%), respectively than *E. melliodora* (77.2%), indicating that their presence and percentages may responsible for the highly insecticidal properties against *P.solenopsis* 3rd instar nymph.

Habibpour *et al.*, (2020) showed that pulegone (51.49%), menthone (22.75%), and 1,8-cineole (11.69%) were the principal components of *Mentha longifolia* (F: Lamiaceae) and menthone (36.51%), menthene (28.51%), menthol (8.12%), and 1, 8-cineole (7.66%) were the principal components of *Mentha piperita* (F: Lamiaceae). **Albazaz and Al-Naqqash (2019)** showed that ɤ-citral represents the major compound in the *Cymbopogon citratus* oil with

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

(42.27%) and this result is comparable to this study **Shah *et al.*, (2011)** result which identified several volatile oil in lemon grass with α-citral being the major compound with (40.8%). The quality of lemon grass oils determines by citral content **(Pengelly 2004** and **Negrelle and Gomes 2007)**. **Breitmaier (2008)** mentioned that terpenoids are volatile substances that provide plants and flowers with their fragrance. They find widely in the leaves and fruits of higher plants, citrus, conifers, and eucalyptus. Our finding is in agreement with those obtained by **Abdelgaleil *et al.*, (2009)** who suggested that AChE may be a target for monoterpenes. Similar results were also documented by **Sharaby and El-Nujiban, (2015), Tayoub *et al.*, (2012), Lee *et al.*, (2001),** and **Lee *et al.*, (2003)** who reported that terpenes such as 1,8-cineole have an insecticidal and repellent activity. Such results were also recorded by **Oladeji *et al.*, (2019)** who obtained that citronella oil (one of the essential oils obtained from the leaves and stems of different types of lemongrass that contains the monoterpene ɤ-citral) is used as an insecticide (bio-pesticide). **Tripathi *et al.* (2009)** indicated that EOs obtained from the plant families, including Asteraceae, Myrtaceae, Apiaceae, Lamiaceae, and Rutaceae have insecticidal activity. **Khater (2012)** reported that bio-insecticides (e.g., EOs) have been posted as an alternative to synthetic insecticide in agriculture and public health sectors.

Atmani-Merabet *et al.* (2018) revealed 39 compounds in GC/MS analysis of *Eucalyptus globulus*, essentially oxygenated monoterpenes (86.01%), monoterpenes (5.74%), monoterpenes alcohols (4.05%), and sesquiterpenes alcohols (2.74%). The main constituents of the oil were 1,8-cineole (78.45 %), o-cymene (2.18 %), isopinocarveole (1.74 %), α-pinene (1.69 %), pinocarvone (1.34%) and veridiflrol (1.31%). **Mostafa *et al.* (2018)** found the most abundant constituents of *Thymus vulgaris* (F: Lamiaceae) to be α-sabinene (16.16%), (-)-β-caryophyllene

ISSN 2537_ 0715
IJSRSD (2021): Volume 4, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

(13.79%), thymol (12.53%), and γ-terpinene (5.47%). The *Mentha longifolia* (F: Lamiaceae) essential oil mainly consisted of piperitenone oxide (15.02%), 4-terpinenol (10.10%) and (-)-cyperene (9.21%). *T. vulgaris* essential oil showed a significant relative percentage of monoterpenes (61.32%) than *M. longifolia* (47.28%), indicating that their presence may responsible for the highly insecticidal properties against *P. solenopsis* adult females. **Dayal *et al.*, (1997)** revealed the presence of 80 compounds. Nine compounds identified are ɤ-pinene (8.34%), β-pinene (4.43%), myrcene (0.24%), terpinene or phellandrene (12.73%), p-cymene (7.25%), limonene (1.97%), cineole (52.11%), borneol (0.22%) and ɤ-terpineol (0.82%).

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

Table (1): Susceptibility of 3rd instar nymphs of cotton mealybug *Phenacoccus solenopsis* (Tinsley) 3rd day after oils application.

Tested compounds	LC_{50} (ppm) its limits at 95%		LC_{90} (ppm) its limits at 95%		Slope ± SE	Toxicity Index %	X^2
Diver® Mineral oil 97%	15080.433		95566.056		1.598 ± 0.505	20.574	1.780
	9635.239	69986.889	32493.492	8334404.943			
Rosemary *Salvia rosmarinus*	**3102.591**		13226.041		2.035 ± 0.600	100	1.279
	2027.288	12726.413	5279.379	380252.691			
Lemon Grass *Cymbopogon citratus*	3323.293		17074.043		1.803 ± 0.559	93.359	4.641
	2046.845	18704.277	5950.704	1148196.442			
Camphor Tree *Eucalyptus melliodora*	4966.815		22656.729		1.944 ± 0.598	62.466	0.484
	2757.552	45341.654	7303.361	2007112.599			

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

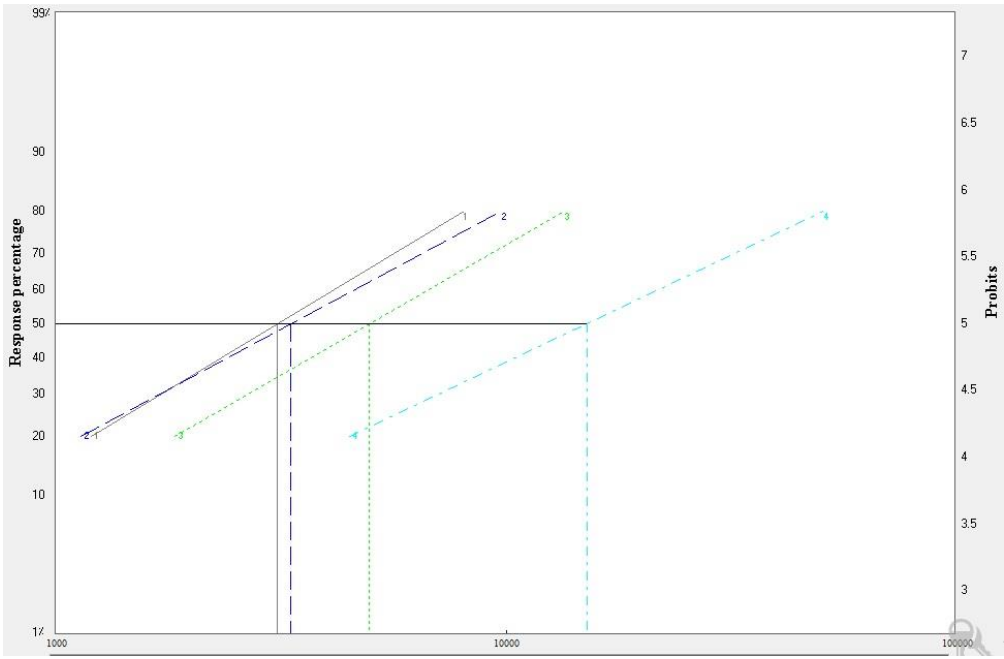

Fig. (1): Susceptibility of 3rd instar nymph of cotton mealybug *Phenacoccus solenopsis* (Tinsley) 3rd day after oils application.

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

Table (2): Susceptibility of 3rd instar nymphs of cotton mealybug *Phenacoccus solenopsis* (Tinsley) week after oils application.

Tested compounds	LC_{50} (ppm) its limits at 95%		LC_{90} (ppm) its limits at 95%		Slope ± SE	Toxicity Index %	X^2
Diver®	1381.831		8974.716		1.577 ± 0.444	49.215	3.457
Mineral oil 97%	258.919	2384.211	6159.096	21760.094			
Rosemary	740.591		4226.784		1.694 ± 0.494	91.828	1.545
Salvia rosmarinus	435.023	981.740	2354.298	31914.664			
Lemon Grass	**680.073**		4020.773		1.660 ± 0.496	100	1.154
Cymbopogon citratus	358.310	908.524	2250.827	32246.064			
Camphor Tree	2483.432		13385.587		1.752 ± 0.533	27.384	3.584
Eucalyptus melliodora	1678.338	9157.025	5090.531	564830.133			

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

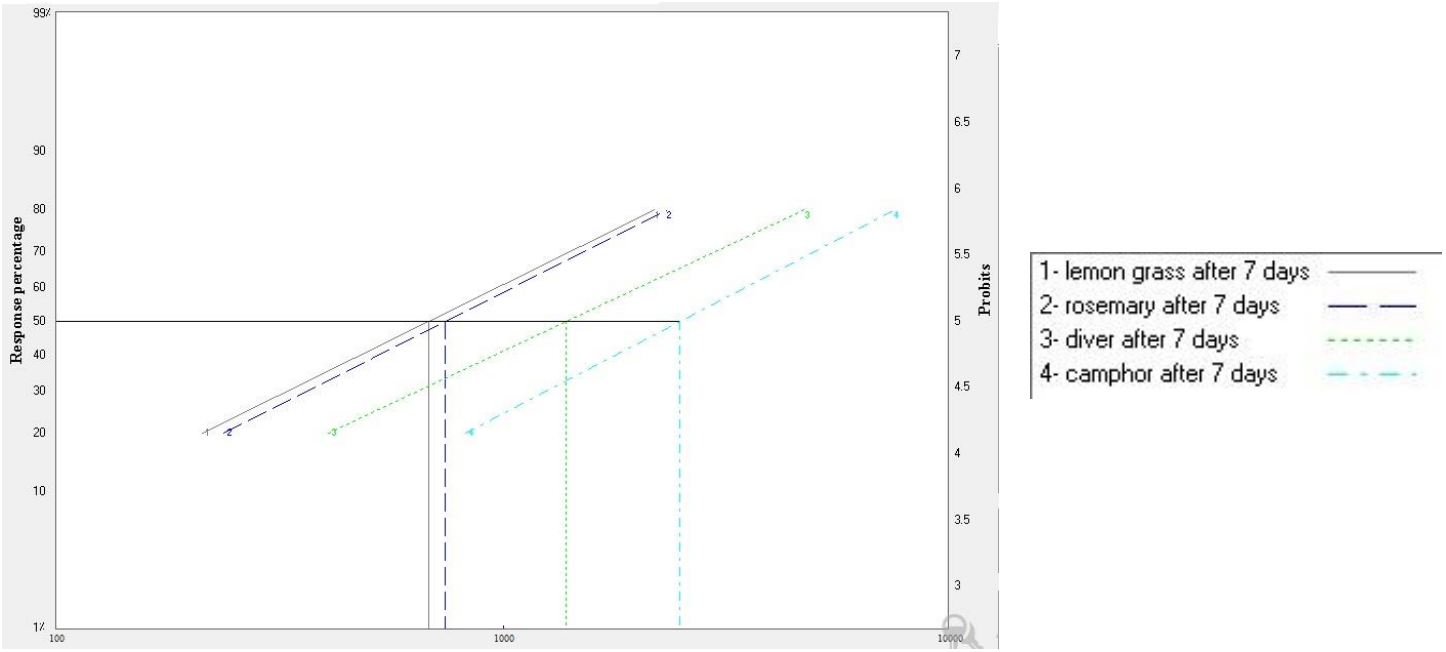

Fig. (2): Susceptibility of 3rd instar nymph of cotton mealybug *Phenacoccus solenopsis* (Tinsley) week after oils application.

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

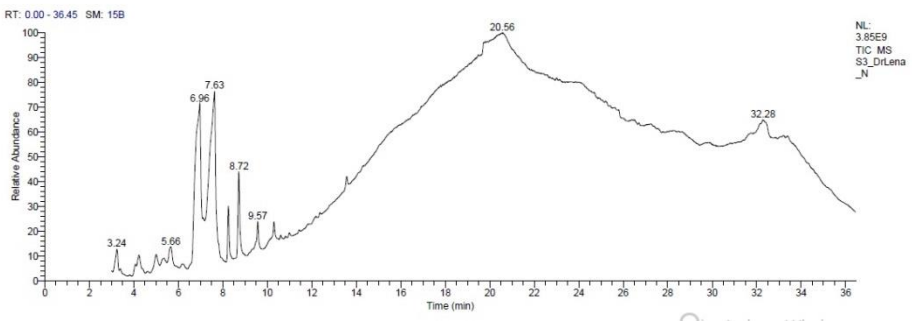

Fig. (3): GC chromatogram of Lemon grass *Cymbopogon citratus*

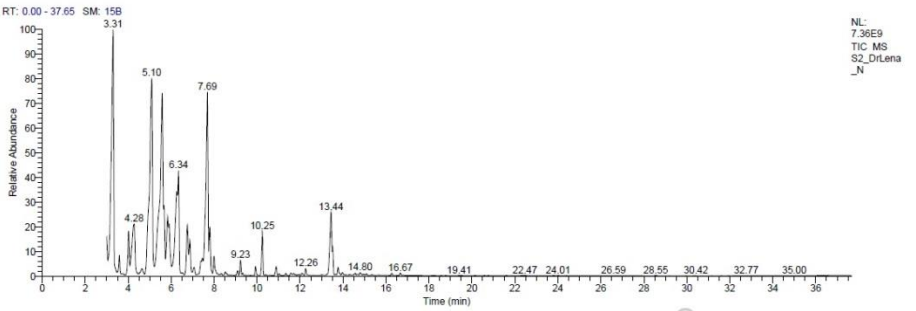

Fig. (4): GC chromatogram of Rosemary *Salvia rosmarinus*

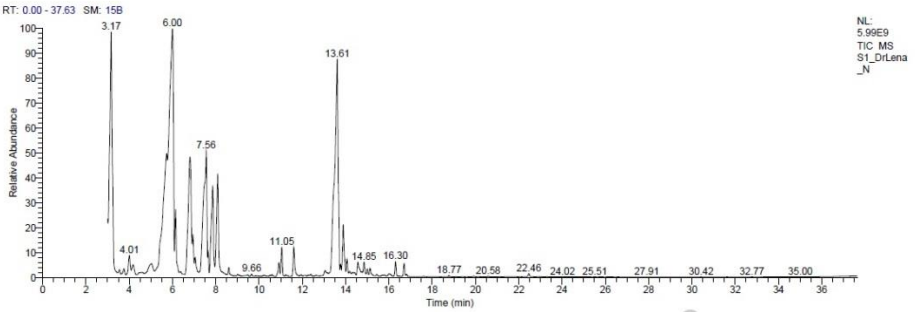

Fig. (5): GC chromatogram of Camphor *Eucalyptus melliodora*

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

Table (3): Chemical constituents of the essential oils identified by GC/MS technique.

No.	Compound name	Retention time (RT.)	Molecular formula (MF)	Molecular weight	*E. melliodora* Area %	*S. rosmarinus* Area %	*C. citratus* Area %
Monoterpenes Hydrocarbons							
1	*p*-cymene	3.18	$C_{10}H_{14}$	134	16.62		
2	ɤ-terpinene	3.59	$C_{10}H_{16}$	136		0.92	
3	*p*-cymenene	4.01	$C_{10}H_{12}$	132	0.90		
4	*p*-mentha-1,4(8)-diene	4.03	$C_{10}H_{16}$	136		2.18	
5	3-isobutyl cyclohexene	5.03	$C_{10}H_{18}$	138	0.89		
6	(+)-limonen	6.76	$C_{10}H_{16}$			3.17	
7	camphene	9.23	$C_{10}H_{16}$	136		0.74	
Total					**18.41**	**7.01**	
Oxygenated Monoterpenes							
8	eucalyptol	3.24	$C_{10}H_{18}O$	154			2.90
9	1,8-cineol	3.31	$C_{10}H_{18}O$	154		18.37	
10	cosmen-2-ol	3.76	$C_{10}H_{16}O$	152	0.39		
11	linalool	4.19	$C_{10}H_{18}O$	154	0.51		1.89
12	3,7-dimethylocta-1,6-dien-3-ol	4.29	$C_{10}H_{18}O$	154		3.93	
13	chrysanthenone	4.64	$C_{10}H_{14}O$	150		0.49	
14	norbornan-2-one	4.91	$C_{10}H_{16}O$	152		0.59	
15	*p*-menth-8-en-2-one	5.00	$C_{10}H_{16}O$	152			1.85
16	norbornan-2-one	5.11	$C_{10}H_{16}O$	152		12.53	
17	*cis*-verbenol	5.35	$C_{10}H_{16}O$	152			1.29
18	isoborneol	5.38	$C_{10}H_{18}O$	154		10.98	
19	terpinen-4-ol	5.42	$C_{10}H_{18}O$	154	2.56	1.98	
20	*trans*-verbenol	5.67	$C_{10}H_{16}O$	152		0.39	2.59
21	4-isopropylcyclohex-2-enone	5.85	$C_9H_{14}O$	138		2.74	
22	(-)-*β*-fenchol	5.91	$C_{10}H_{18}O$	154		2.21	
23	(z)-tagetenone	6.01	$C_{10}H_{14}O$	150	20.56		
24	(-)-cis-Sabinol	6.15	$C_{10}H_{16}O$	152	2.06		
25	levoverbenone	6.27	$C_{10}H_{14}O$	150		10.73	
26	*p*-isopropyl benzaldehyde	6.80	$C_{10}H_{12}O$	148	7.99		
27	trans-shisool	6.88	$C_{10}H_{18}O$	154		2.27	
28	*p*-menth-1(7)-en-2-one	6.95	$C_{10}H_{16}O$	152	0.68		
29	ɤ-citral	6.97	$C_{10}H_{16}O$	152			70.44
30	*p*-menth-1-en-3-one	7.06	$C_{10}H_{16}O$	152	0.43		
31	geraniol	7.09	$C_{10}H_{18}O$	154		0.43	
32	phellandral	7.45	$C_{10}H_{16}O$	152	11.82	0.28	
33	2-caren-10-al	7.65	$C_{10}H_{14}O$	150	0.48		
34	bornyl acetate	7.70	$C_{12}H_{20}O_2$	196		13.02	
35	cumic alcohol	7.81	$C_{10}H_{14}O$	150		1.89	

أكاديمية البحث العلمي والتكنولوجيا
Academy of Scientific Research & Technology

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

36	*p*-cymen-7-ol	7.87	$C_{10}H_{14}O$	150	5.16		
37	carvacrol	8.10	$C_{10}H_{14}O$	150	5.83	1.03	
38	neryl acetal	8.25	$C_{12}H_{22}O_2$	198			5.12
39	hexahydro-3-methylenebenzofuran-2(3H)-one	8.60	$C_9H_{12}O_2$	152	0.32		
40	geranyl vinyl ether	9.57	$C_{12}H_{20}O$	180			2.98
Total					**58.79**	**83.86**	**89.06**
Total Monoterpenes					**77.2**	**90.87**	**89.06**
Sesquiterpenes Hydrocarbons							
41	caryophyllene	10.25	$C_{15}H_{24}$	204		2.45	
42	humulene	10.89	$C_{15}H_{24}$	204		0.52	
43	aromadendrene	11.06	$C_{15}H_{24}$	204	1.26		
44	cadina-1(10),4-diene	12.26	$C_{15}H_{24}$	204		0.35	
Total					**1.26**	**3.32**	
Oxygenated Sesquiterpenes							
45	(+)-cycloisolongifol-5-ol	10.92	$C_{15}H_{24}O$	220	0.68		
46	(-)-spathulenol	13.43	$C_{15}H_{24}O$	220	14.60	5.79	
47	viridiflorol	13.77	$C_{15}H_{26}O$	222	0.24		
48	ledene oxide-(ii)	13.90	$C_{15}H_{24}O$	220	2.73		
49	(1R,7S)-Germacra-4(15),5,10(14)-trien-1β-ol	14.85	$C_{15}H_{24}O$	220	0.50		
50	ylangenal	16.31	$C_{15}H_{22}O$	218	1.21		
Total					**19.96**	**5.79**	
Total Sesquiterpenes					**21.22**	**9.11**	
51	γ-dodecalactone	8.72	$C_{12}H_{22}O_2$	198			9.22
52	2-(7-heptadecynyloxy) tetrahydro-2H-Pyran	10.30	$C_{22}H_{40}O_2$	336			1.72
53	methyleugenol	9.93	$C_{11}H_{14}O_2$	178		0.46	
Total						**0.46**	**10.94**
Total					**98.42**	**100.44**	**100.00**

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

CONCLUSION

Phytochemicals (biopesticides based on essential oils) appeared to be a complementary or alternative method in crop production and integrated pest management. Concerning the development of quality herbal medicine adjustment of the extracts, phytopharmacology of altered extracts, isolation, and representation of active phytopharmaceuticals, illumination of the mechanism of action of the isolated compounds and clinical features of the compounds are much required to develop new natural products which are more efficient, but, above all, will overcome the problem of increasing pest resistance.

REFERENCES

Abdelgaleil, S.A.M.; Mohamed, M.I.E.; Badawy, M.E.I. and El-arami, S.A.A. (2009). Fumigant and contact toxicities of Monoterpenes to *Sitophilus oryzae* (L.) and *Tribolium castaneum* (Herbst) and their inhibitory effects on acetylcholinesterase activity. Journal of Chemical Ecology. 35:518-525.

Abro, G.H.; Syed, T.S.; Tnuio, G.M. and Khuro, M.A. (2004). Performance of transgenic Bt. cotton against insect pest infestation. Biotechnol. 3:75-81.

Ahmad F.; Akram, W.; Sajjad, A. and Imran, A. (2011). Management practices against cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae). International Journal of Agricultural and Biology. 13(4):547–552.

Alagarmalai, J. (2017). Phytochemicals: As Alternate to Chemical Pesticides for Insects Pest Management. Current trends in biomedical engineering & biosciences. Volume 4, issue 1.

Alakhdar,H.H.; Daar, R.A.A. and Abd-Elrahman, T.A. (2021). Toxicological evaluation and residual analysis of some acaricides against two-spotted spider mite *Tetranychus urticae* by using certain ground spraying equipment on cotton and its intercrops in Egypt. International Journal of Entomology Research. 6 (1): 58-67.

Alakhdar, H.H. and Ghareeb, Z.E. (2021). Relative toxicity of two natural compounds compared to abamectin against some soybean pests under period rates. OCL, 28, 32. https://doi.org/10.1051/ocl/2021018

Al-Naqqash, H. and Albazaz, Z. (2019). GC-Mass and Phytochemical Investigation of *Cymbopogon citratus*. Research Journal of Pharmacy and Technology. 12(1):67.

Arif, M.; Gogi, M.D.; Arfat, A.; Anjum, S.; Zain-ul, A.; Waqas, W. and Ahmad, N. (2012). Host-plants mediated population dynamics of cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) and its parasitoid, *Aenasius bambawalei* Hayat (Hymenoptera: Encyrtidae). Pakistan Entomologist. 34(2):179-184.

Arif, M.I.; Rafiq, M. and Ghaffar, A. (2009). Host plants of cotton mealybug (*Phenacoccus solenopsis*): a new menace to cotton agroecosystem of Punjab, Pakistan. International Journal of Agricultural and Biology. 11(2):163–167.

Atmani-Merabet, G.; Belkhiri, A.; Dems, M.A.; Lalaouna, A.; Zakaria Khalfaoui, Z. and Mosbah, B. (2018). Chemical composition, toxicity, and acaricidal activity of *Eucalyptus globulus* essential oil from Algeria. Curr. Issues Pharm. Med. Sci., Vol. 31, No. 2, Pages xx-xx.

Breitmaier, E. (2008). In: Terpenes: Flavors, Fragrances, Pharmaca, Pheromones, Wiley-VCH Verlag GmbH & Co. KGaA, Germany, 1st Edition.

Choi, W.; Lee, E.; Choi, B.; Park, H. and Ahn, Y. (2003). Toxicity of Plant Essential Oils to *Trialeurodes vaporariorum* (Homoptera: Aleyrodidae). J. Econ. Entomol. 96(5):1479-1484.

Culik, M.P. and Gullan, P.J. (2005). A new pest of tomato and other records of mealybugs (Hemiptera: Pseudococcidae) from Espirito Santo, Brazil. Zootaxa. 964:1–8.

Dayal, R.; Parcha, V. and Bartley, J.P. (1997). GC-Mass studies of oil from Eucalyptus hybrid leaves. Indian Perfumer. 41(4). PP 151-153.

Delvin, J.F. and Zettel, T. (1999). Ecoagriculture: Initiatives in Eastern and Southern Africa. Weaver Press, Harare.

Dhawan, A.K.; Sing, J. and Sidhu, A.S. (1980). *Maconellicoccus* sp. attacking Arboreum cotton in Punjab. Journal of Science and Culture. 46(7):258.

Dinesh, K.P.; Sachin, M.; Veena, B.J.; Seetharama, H.G.; Sreedharan, K. and Kumar, P.K. (2003). Evaluation of botanicals against mealybug *Planococcus citri* Risso and its effect on parasitoid and attendant ant. Journal of Coffee Research. 31(2):139–152.

El-Sonbaty, S.M.; Abasse, A.E.; Abd-El Wahab, H.A.; Naema, A.A.; Mohammed, E. and Mohammed, M. (2018). Nano Bioinsecticides based on Essential oils Against *Phenacoccus solenopsis*. Egyptian Academic Journal of Biological Sciences A Entomology. 11(5):1-12.

Gross, S.; Gefen, D.; Rotman, N.; Tadmor, U.; Zemer, B.; Gotlib, A. and Gefen, Y. (2001). Chemical control of spherical mealybug (*Nipaecoccus viridis*) (Newstead) in citrus. Alon Hanotea. 54(6):234–240.

Habibpour, B.; Roozdar, E.; Mossadegh, M.S. and Mahmoodi, M. (2020). Efficacy of different formulations of essential oils against the cotton mealybug, *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae) under laboratory conditions. Moharramipour Journal of Entomological Society of Iran. 40(2):183-199.

Hargreaves, H. (1948). List of recorded cotton insects in the world. Harrisson and Sons. Commonwealth Inst. Ento., London. pp.50.

Hayat, B.; Farman, U.; Abid Farid, P.A. and Neil, C. (2015). Toxicity of Neem seed *Azadirachta indica* Juss (Meliaceae) different solvents extracts against cotton mealybug *Phenacoccus solenopsis* Tinsley (Sternorrhyncha: Pseudococcidae) under Laboratory conditions. Journal of Entomology and Zoology Studies. 3(4),45-49.

Khater, H.F. (2012). Prospects of botanical biopesticides in insect pest management. Pharmacologia, 3: 641-656.

Kostyukovsky, M.; Rafaeli, A.; Gileadi, C.; Demchenko, N. and Shaaya, E. (2002). Activation of octopaminergic receptors by essential oil constituents

isolated from aromatic plants: possible mode of action against insect pests. Pest Management Science. 58(11):1101-1106.

Lee, B.H.; Choi, W.S.; Lee, S.E. and Park, B.S. (2001). Fumigant toxicity of essential oils and their constituent compounds towards the rice weevil, *Sitophilus oryzae* (L.). Crop Prot., 20:317-320.

Lee, S.; Peterson, C.J. and Coats, J.R. (2003). Fumigation toxicity of monoterpenoids to several stored product insects. J. Stored Prod. Res., 39: 77-85.

Li, X.W.; Lu, X.X.; Zhang, Z.J.; Huang, J.; Zhang, J.M.; Wang, L.K.; Hafeez, M.; Fernández-Grandon, G.M. and Lu, Y.B. (2021). Intercropping Rosemary (*Rosmarinus officinalis*) with Sweet Pepper (*Capsicum annum*) Reduces Major Pest Population Densities without Impacting Natural Enemy Populations. Insects 2021. 12:74.

Manjunath, T.M. (2004). Bt cotton in India. In: The technology wins as the controversy wanes. Available at: www. Monsanto.co.uk / news ukshowlib.html? wid = 8478.

Masango, P. (2005). Cleaner production of essential oils by steam distillation. J. Cleaner Prod. 13:833-839.

Mohamed, E.M.; Naglaa, M.Y. and Anwaar, M.A. (2018). Insecticidal activity and chemical composition of plant essential oils against cotton mealybug, *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcidae). Journal of Entomology and Zoology Studies. 6(2), 539-543.

Mohyuddin, A.L.; Jillani, G.; Khan, A.G.; Hamza, A.; Ahamad, I. and Mahmood, Z. (1997). Integrated pest management of cotton pests by conservation, redistribution and augmentation of natural enemies in Pakistan. Pakistan Journal of Zoology. 29:393–398.

Mossa, A.H. (2016). Green Pesticides: Essential oils as biopesticides in insect-pest management. Journal of Environmental Science and Technology. 9(5):354-378.

Mostafa, M.E.; Youssef, N.M. and Abaza, A.M. (2018). Insecticidal activity and chemical composition of plant essential oils against cotton mealybug, *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcidae). Journal of entomology and zoology studies. 6(2):539-543.

Muhammad, I.U.; Syed, M.A.Z.; Muhammad, A.; Yasir, I.; Samina, K.; Jaime, M.O. and Muhammad, N. (2017). Toxicity of Botanicals and Conventional Insecticides to *Aenasius bambawalei* Hayat, an Endoparasitoid of Cotton Mealybug, *Phenacoccus solenopsis* Tinsley. Southwestern Entomologist. 42(4):941-952.

Negrelle, R.R.B. and Gomes, E.C. (2007). *Cymbopogon citratus* (DC.) Stapf: chemical composition and biological activities. Revista Brasileira de Plantas Medicinais. 9:80-92.

Oladeji, O.S.; Adelowo, F.E.; Abraham, O.K.; Ayodele, D.; Bankole, D. and Oladipo, C. (2019). Phytochemistry and Pharmacology of Cymbopogon citratus: A review. Scientific African. 6:e00137.

Pengelly, A. (2004). The Constituents of medicinal plants (Eds): An Introduction to the chemistry and therapeutics of herbal medicine. United Kingdom: CABI Publishing. pp. 85-103.

Peschiutta, M.L.; Pizzolitto, R.P.; Ordano, M.A.; Zaio, Y.P. and Zygadlo, J.A. (2017). Laboratory evaluation of insecticidal activity of plant essential oils against the vine mealybug *Planococcus ficus*. 56:79-83.

Priestley, C.M.; Williamson, E.M.; Wafford, K.A. and Sattelle, D.B. (2003). Thymol, a constituent of thyme essential oil, is a positive allosteric modulatorof human GABA receptors and a homo-oligomeric GABA receptor from *Drosophila melanogaster*. British Journal of Pharmacology. 140(8):1363-1372.

Prishanthini, M. & Vinobaba, M. (2014). Efficacy of some selected botanical extracts against the cotton mealybug *Phenacoccus solenopsis* (Tinsley) (Hemiptera: Pseudococcidae). International Journal of Scientific and Research Publications. 4:1-6.

Puntener, W., (1981). Manual for field trials in plant protection second edition.Agri. Divis. Ciba-Geigy Limited.

Rashid, H.; Polyak, L. and Mosley-Thompson, E. (2011). Abrupt Climate Change: Mechanisms, Patterns, and Impacts. Journal of Geophysical Monograph Series. American Geophysical Union, Washington D.C. 193:2. Doi:10.1029/GM193. Issn: 0065-8448.

Robert, A. (1995). Text book of identification of essential oil components gas chromatography/ mass spectroscopy. 2nd ed. 46:1460.

Saeed, S.; Ahmad, M. and Kown, Y.J. (2007). Insecticidal control of the mealybug *Phenacoccus gossypiphilous* (Hemiptera: Pseudococcidae), a new pest of cotton in Pakistan. Entomological Research. 37 (2):76–80.

Sahito, H.A.; Abro, G.H.; Khuhro, R.D. and Buriro, A.S. (2009). Varietal Resistance of Cotton Crop against Mealybug, *Phenacoccus solenopsis* Tinsley. Pak. Jr., Agri., Agri. Engg. Vet. Sci. 25(1):34-38.

Saini, R.K.; Sharma, S.S. and Rohilla, H.R. (2009). Mealybug *Phenacoccus solenopsis* Tinsley and its survival in cotton ecosystem in Haryana. In: Proceedings of National Symposium on Bt-cotton: opportunities and prospectus. Central Institute of Cotton Results, Nagpur, Indie. 150 pp.

Satpute, M.T.; Rizvi, N.H. and Nizamani, S.M. (1998). Some factors affecting cotton aphid, whitefly, bollworms infestation and cotton yield. Ann. Agri. Sci. 39: 431-439.

Schneider-Orelli, O., (1947). Entomologisches Pratikum. HR Sauerlander and Co, Aarau.

Shah, G.; Shri, R.; Panchal, V.; Sharma, N.; Singh, B. and Mann, A.S. (2011). Scientific basis for the therapeutic use of *Cymbopogon citratus*, stapf (Lemon grass). J. Adv. Pharmac. Technol. Res. 2(1):3-8.

Sharaby, A. and El-Nujiban, A. (2015). Adverse effect of pure terpenes and some combinations against the black cutworm, *Agrotis ipsilon* (Hufn.) (Lepidoptera: Noctuidae). Egypt. J. Biol. Pest Control, 25: 401-406.

Sun, Y.P. (1950). Toxicity index an improved method of comparing the relative toxicity of insecticides. J. Econ. Ent. 43:45-53.

Sunitha, N.D.; Jagginavar, S.B. and Biradar, A.P. (2009). Bioefficacy botanicals and newer insecticides against grape vine mealybug, *Maconellicoccus hirsutus* (Green). Karnataka Journal of Agricultural Sciences. 22 (3):710–711.

Suresh, S.; Jothimani, R.; Sivasubrmanian, P.; Karuppuchamy, P.; Samiyappan, R. and Jonatha, E.I. (2010). Invasive mealybugs of Tamil Nadu and their management. Karnataka Journal of Agricultural Sciences. 23 (1):6–9.

Tanwar, R.K.; Jeyakumar, P. and Monga, D. (2007). Mealybugs and their management. National Centre for Integrated Pest Management. LBS Building. Pusa Campus. New Delhi, India. Tech. Bull. No. 19. 12 pp.

Tayoub, G.; Odeh, A. and Ghanem, I. (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 Polonica, 58: 26-37.

Tripathi, A.K.; Upadhyay, S.; Bhuiyan, M. and Bhattacharya, P.B. (2009). A review on prospects of essential oils as biopesticide in insect-pest management. J. Pharmacogn. Phytother. 1: 52-63.

Zellner, D.A.; Dugo, P.; Dugo, G. and Mondello, L. (2010). Analysis of Essential Oils. In: Essential Oils Science, Technology and Applications, Baser, C., K. Husnu and G. Buchbauer (Eds.). CRC Press, New York, pp:151-177.

ISSN 2537_ 0715
IJSRSD (2021): Volume 1, Issue 1, June 2021
Received: April 2021, Accepted: June 2021

International Journal of Scientific
Research and Sustainable Development

الزيوت العطرية كمبيدات حشرية خضراء: تحليل GC/MS ودراسات سمية علي آفة بق القطن الدقيقي *Phenacoccus solenopsis* (Tinsley) (نصفية الأجنحة – المكورات الكاذبة)

لينا حسن الراضى على سعد[1]، سلوى السعيد نجم[2]، عادل عبد المنعم صالح[2]، أحمد السيد محمود عبد المجيد[1]

[1] معهد بحوث وقاية النباتات – مركز البحوث الزراعية – الدقى – الجيزة – مصر

[2] كلية الزراعة – جامعة المنصورة

أجريت هذه الدراسة بهدف تقييم سمية ثلاثة زيوت عطرية مستخلصة من أوراق نبات الروزماري، نبات حشيشة الليمون ونبات الكافور بالمقارنة مع الزيت المعدني دايفر تحت الظروف المعملية علي العمر الثالث لحورية حشرة بق القطن الدقيقي مع توضيح التركيب الكيميائي للزيوت العطرية. أوضحت النتائج، أبرز الزيوت العطرية سمية علي العمر الثالث لحورية بق القطن الدقيقي بعد مرور ثلاثة أيام من المعاملة كان زيت الروزماري يليه زيت حشيشة الليمون. فكانت قيمة التركيز القاتل لـ 50% من الأفراد 3102,591 و3323,293 جزء في المليون، علي التوالي؛ وكانت قيمة التركيز القاتل لـ 50% من الأفراد بعد مرور سبعة أيام لكل من زيت حشيشة الليمون يليه زيت الروزماري 680,073 و 740,591 جزء في المليون، علي التوالي. تم استخدام مطياف الكتلة اللوني لتحليل الزيوت العطرية ولتحديد العناصر الأكثر نشاطا. أوضحت النتائج أن أكثر المكونات المكونة لزيت الكافور هو (زد)-تاجيتينون (20,56%) و بيسايمن (16,62%). وكان زيت الروزماري يتركب من 8,1سينول (18,37%)، بورنيل أسيتات (13,02%) و نوربورنان-2-وان (12,53%)، وكان المكون الرئيسي لزيت حشيشة الليمون هو ألفا-سيترال (70,44%). أظهرت الزيوت العطرية لكل من نبات الروزماري وحشيشة الليمون وجود نسبة عالية معنوية من التربينات الأحادية (90,87% - 89,06%)، علي التوالي، مما يشير إلي أن هذه النسب قد تكون المسؤلة عن درجة السمية المرتفعة ضد آفة بق القطن الدقيقي.

الكلمات الدالة: زيوت عطرية – بق القطن الدقيقي – الروزماري – حشيشة الليمون – الكافور - مطياف الكتلة اللوني.