

BIOCHEMICAL CHARACTERIZATION AND INSECTICIDAL ACTIVITY OF DIFFERENT SOLVENT CRUDE EXTRACTS OF *LANTANA* *CAMARA* L. ON DIAMONDBACK MOTH, *PLUTELLA XYLOSTELLA* (LINN.)

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Abstract

A study was made to evaluate the insecticidal action of *Lantana camara* L. against diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae). Sixty to seventy five grams of ground plant material were successively extracted using eight organic solvents viz., acetone, benzene, chloroform, ethanol, ethyl acetate, petroleum ether, hexane, and methanol in Soxhlet apparatus for 24 hrs and tested against *P. xylostella* at six concentrations (1, 2, 4, 6, 8 and 10 per cent). Bioassay results indicated that the toxicity increased proportional to the concentrations of the all extracts. Among the extracts of different solvents experimented, hexane extract proved to be the best solvent followed by others; we also have compared these extracts with neem oil of same concentrations. In all solvent extracts eight and ten per cent concentration of *L. camara* had stronger ovicidal and oviposition deterrent effects with low larvicidal activities. Plant metabolites are highly diverse, having distinct functions according to their structure and Gas chromatography Mass Spectrometry (GC-MS) analysis of hexane extract revealed the presence of 19 major phytochemicals including caryophyllene, caryophyllene oxide, 2-hexadecen-1-ol, benzene, hexatriacontane, tetrapentacontane, 1, 3-cyclohexadiene-1-carboxaldehyde, 6S-2,3,8,8-tetramethyltricyclo[5.2.2.0(1,6)]undec-2-ene, 3-nonanone, phytol and squalene etc.

Keywords: *Lantana camara*, GC-MS analysis, contact toxicity, ovicidal effect

Introduction

Cabbage and cauliflower are important cash crops for farmers often produced under small holder conditions throughout tropical and subtropical areas of Asia, Africa, Latin America and the Caribbean countries. In India, vegetables play important role in nutritional security, economic viability and source of remunerative income for many small and marginal farmers under intensive farming system. The cole vegetables are cultivated in 4.00 and 4.34 lakh ha producing 9039.00 and 8573.00 MT with an average yield was 22.6 and 19.80 t ha⁻¹ of cabbage and cauliflower respectively in India. During 2013-2014, India produced 162.19 million tonnes of vegetables and exported worth of Rs.

5462.93 crores (Indian Horticulture Database, 2013).

The production share of cruciferous vegetable crops (2013-14) was to the extent of 5.5 and 5.3 per cent of cabbage and cauliflower respectively and the yield loss estimated upto 17- 99 per cent of both (Uijtewaal, 2006; Uthamasamy *et al.* 2011 and IHD, 2013). In Tamil Nadu, it occupies an area of 24000 and 9500 ha with an annual production of 130.42 and 209.17 MT and productivity is 50 and 22T ha⁻¹ of cabbage and cauliflower respectively. The economic loss due to this pest has been estimated worldwide to be US\$ 4-5 billion (Zalucki *et al.*, 2012) and US\$ 16 million annually in India (Mohan and Gujar, 2003). Several

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applications of synthetic insecticides on crucifers are common among commercial farmers (FAO, 2005). *Lantana* (Verbenaceae) is mostly distributed in approximately 50 countries in the world. Introductions of non-native or agricultural plants into novel habitats without their co-evolved counterparts may induce novel plant-insect interactions (Strong, 1979).

Many kinds of monoterpenes from plant sources have been evaluated as feeding deterrents against insects (Koul, 1982). The major secondary substances found among plants are organic compounds such as alkaloids, terpenes, saponins, phenol, cardiac and cyanogenic glycosides, nitro-containing compounds, resins and certain proteins and acids (Lewis and Elvin-Levis, 1977). Oil extracted from *L. camara* are known to exhibit ovicidal, insecticidal, antifeedant, attractant, repellent, antiviral and anti-juvenile hormone activities (Rejesus, 1986). However, information on the efficacy of *L. camara* on cruciferous vegetables is very scarce. Keeping these in view, the present study was made to evaluate the efficacy of different solvent crude extracts of *L. camara* against diamondback moth *P. xylostella* in comparison with commonly used botanical neem oil.

Materials and Methods

The study focused on the effectiveness of different organic solvent extractions from the aerial part of *L. camara* tested against *P. xylostella* in cole vegetables. Experiments were conducted at Insectary, Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore, from August 2014 to December 2014.

Collection of test plant materials

Aerial parts of the test plant *L. camara* were collected from Thondamuthur and Aalandurai block of Coimbatore district during November 2013. The samples were air-dried for 15-20 days under shade. After complete shade drying, the plant parts were pulverized into powder with the help of local motor grinder. The plant material was extracted by Soxhlet extraction method. The aqueous extract of these plant materials was prepared on per cent basis as per Sharma *et al.* (1997) for which a stock solution of 10 per cent

concentration was prepared by dissolving 10 gram of plant material in 90 ml of distilled water and used at different concentrations.

Soxhlet Extraction

The plant material was subjected to Soxhlet extraction suggested by Sukthamrong *et al.* (1981) and Sharma and Gupta (2009) in order to extract more active principles. Known amount (50 - 75g) of plant material of each solvent was filled into the Soxhlet apparatus. A cotton plug was used at the place of thimble to stop the entry of the crude material into the siphoning tube. The required organic solvents *viz.*, acetone, benzene, chloroform, ethanol, ethyl acetate, petroleum ether, hexane, and methanol were filled up five times more than total amount of the sample material into the flask of the apparatus. The apparatus was then connected with the water supply to the condenser. The temperature of the heating mantle was maintained according to the boiling point of organic solvents. The process was carried out for 24 hrs for each sample of the solvent.

The pooled extract was then filtered using a Whatmann filter paper no.1 and concentrated by rotary evaporation at 40°C. After drying in desicator, crude extracts were weighed, stored in stock vials and kept in refrigerator (0 - 4°C) for further use. It was further purified by flash or column chromatography (anhydrous Na₂SO₄ + silica gel + dehydrated charcoal) for GC MS analysis.

Gas Chromatography – Mass Spectrometry analysis of *L. Camara* leaf extract

A shimadzu QP – 2010 plus GC-MS was used in pesticide Toxicology laboratory, Department of Agricultural Entomology and method described by Shettima *et al.* (2013). The GC-MS was equipped with a split injector and an ion-trap mass with 220°C spectrometer detector together with a fused silica capillary column (RXI-1MS) having a thickness of 0.25µm, dimensions of 30m x 0.25mm and temperature limits of 60°C to 260°C. The column temperature was programmed between 60°C and 260°C at a rate of 3.0ml/min. The mass range and temperature of the

injector and detector were at 500 M/z of 220°C and 200°C respectively. Helium gas was used as a carrier gas at a flow rate of 0.95 ml per minute.

Laboratory rearing of the diamondback moth, *Plutella xylostella* (L.)

Raising of mustard seedlings

The soil mixture required for raising mustard seedlings were prepared by mixing soil, compost and leaf litter at a ratio of 1:1:1. Plastic tea cups (6 x 3cm) with holes at the bottom were filled with soil mixture to a height of 2.5 cm. Mustard seeds, treated with carbendazim were sown evenly over the entire soil surface and watering was done once in two days. The seeds germinated in about three days at a room temperature of 28±5°C. The seedlings, at a height of 4 - 5 cm, were used for oviposition by female DBM.

Adult oviposition and collection of eggs

The DBM culture was initiated with the larvae collected from the field of Aalandurai Narasipuram and Thondamuthur areas of Coimbatore. The collected larvae were reared on young cauliflower leaves. When they pupated, 50 nos. were transferred to adult emergence cages (30x30x30 cm). Sugar solution (10%) fortified with multi-vitamin drops was provided as adult diet for the moths. A day after the emergence of adults, mustard seedlings were provided for oviposition. The moths laid eggs on both the surfaces of mustard leaves as well as on petioles. Fresh seedlings were provided once in two days until all the adults died.

DBM larval rearing

Larval rearing was carried out in cages with the size of 30x30x30 cm. The first instar larvae hatched in about 3 to 4 days were initially fed by mining into the mustard leaves and later on the entire leaves. For second instar larvae, tender cauliflower leaves were provided as feed material. Most of the larvae migrated to cauliflower leaves within a day and the larvae were provided with fresh leaves every day. To meet the daily requirement of leaves, cauliflower plants were grown continuously

in pots and field. The larval stage lasted for 12 to 14 days and pupation mostly occurred on the lower surfaces of the leaves. The larvae pupated during different dates were collected by using a camel hair brush. To synchronise the emergence of moths, the collected pupae were stored in a refrigerator. When all the larvae were pupated, they were taken out from the refrigerator and kept in the adult emergence cage. The pupal period lasted for 5 to 6 days. DBM were rearing under laboratory conditions at a photoperiod of 12:12 (light: dark) and temperature of 28±4.0°C with RH of 65±5 per cent.

Bioassays (No Choice Test)

Larval contact toxicity

The contact toxicity of *L. camara* crude extracts was evaluated using Potter's tower (Potter, 1952). One ml of each concentration (1, 2, 4, 6, 8 and 10%) was delivered on the ten larvae were placed in a petri plates per replication including the untreated leaf disc. The crude extracts were sprayed through a Potter tower (Burkard, Rickmansworth, UK) on cauliflower leaf discs (9.0 cm diameter) at 0.34 bar (34 kPa) pressure with APSA 80 0.1 ml + one ml spray aliquot. The treatment using organic solvent + APSA 80 0.1 ml served as the control of respective extraction. After complete evaporation, the leaves were transferred to clean bioassay containers over a moistened filter paper. The leaf discs were placed slantingly to rest on side of the container so that larvae can move on either side. Ten larvae were released in each petri dish and three replicates were maintained per treatment. Per cent mortality was observed 24, 48 and 72 h after treatment. All the experiments were carried out in a room temperature with a photoperiod of 12:12 (L: D) and experiments with control mortality more than 20 per cent were discarded and repeated. The mortality data were recorded at different interval till adult emergence. The total per cent mortality data were calculated and corrected using Abbott's formula (Abbott, 1925).

Pupal contact toxicity

The contact toxicity of *L. camara* crude extracts in pupae was evaluated by direct dip bioassay described by Idris and Grafius (1993). About fifty pupae used in each concentration of *P. xylostella* were placed in a 4 cm tea filter and dipped in the defined time for 30 sec. In the control treatment the test insect were tipped in tap water. The pupae were removed and blotted dry on filter paper and transformed onto untreated cabbage leaf discs in 9 cm petri dishes. The mortalities were recorded every 24 hrs upto seven days or died as dark pupae and all the experiments were replicated three times.

Ovicidal toxicity

Ovicidal toxicity was evolved as per Kumar *et al.* (2009). 6 - 8 week old cabbage/ cauliflower seedlings were placed in a screened insect cage (40 x 40 x 40 cm) where 20 pairs of 2 day old moths were placed; after 24 h, the eggs deposited on the seedlings were collected and counted. Fifty eggs deposited on the leaves were dipped into 1 ml of each solution of six concentrations (1, 2, 4, 6, 8 and 10%) for 30 sec and placed in a petri dish. The number of unhatched eggs was counted on the fifth day after treatment. Each treatment, including the control, was replicated three times.

Oviposition deterrent activity

Oviposition deterrent activity was evaluated as per Soontorn and Rejesus (2005). *L. camara* crude extracts of 1, 2, 4, 6, 8 and 10 per cent were sprayed for 6 - 8 week old cabbage and cauliflower seedlings, which were then placed inside a cage. Three seedlings represented in each treatment, including a control that was sprayed with distilled water. Adult *P. xylostella* females were tested for oviposition in a no-choice test on treated and untreated cauliflower plants in a screened cage (30 x 30 x 30 cm) kept in a laboratory at 25°C with 60-70 per cent R.H. For each replicate, mated females (within 24 h) was released and eggs on each leaf per live plant were counted after 24 hrs and mean counts expressed in oviposition deterrent indices (ODI) as: Oviposition

deterency index = $(C-T)/(C+T) \times 100$; where C = number of eggs on control plants and T = number of eggs on treated plants.

Statistical analysis

The laboratory experiment was conducted in completely randomized design. The raw data were subjected to square root transformation and the data on percentage were transformed into arc sine values before statistical analysis. Observed mortality data were converted to percentage and were subjected to probit analysis (Finney, 1971) for obtaining regression equations for dosage mortality response and to determine the LC50 and LC95 values. The mean values were separated using LSD through ANOVA.

Results and Discussion

The active principles from aerial parts of *L. camara* were extracted using eight solvents of different polarity. Among all the solvents used, extracts from petroleum ether, hexane, methanol and chloroform retain their natural appearance. The amount, temperatures of extraction and appearance of each solvent fraction colours are presented in (Table 1). Phytochemical compounds are believed to deter invertebrates from plants, either by acting as antifeedants or by being toxic through hormonal disruption upon ingestion. Most of plants were reported to be insecticidal without specifying the type of action. In these contexts, this experiment assumed importance that the results would compartmentalize toxicity either as contact and/or stomach poisons.

Larval and pupal contact toxicity

The toxicity of different solvents extracts of *L. camara* and neem oil at every 24 hrs is illustrated in (Figures 2 a - j). Significant differences were found among the treatments ($P < 0.05$); and corresponding probit curves/mortality of second instars are represented in (Figure 1). Among the eight solvent crude extracts tested, hexane extract showed greater performance in terms of contact toxicity of larval and pupae, ovicidal toxicity, oviposition deterrent activity as it are evident

from the data. On the basis of probit analysis (Table 2) the efficiency (LC₅₀ and LC₉₅) of extracts was as follows: hexane > chloroform> methanol> ethyl acetate> acetone> petroleum ether> ethanol> benzene.

Among the larval stages second and third instars are highly vulnerable rather than fourth instar and pupal stage Figure 5.

Table 1 Physical characteristics of fractions obtained from leaf of *Lantana camara* L.

S.No.	Organic Solvents and their quantity (Lit.)	Temp. (°C)	Sample Weight(g)		Texture	Colour	
			Leaf	Crude			
1	Acetone	2.0	55-57	270	30.00	Jelly	Dark green
2	Benzene	1.0	80.10	230	30.00	Gummy	Light yellow
3	Chloroform	1.8	60-62	400	30.00	Gummy	Green
4	Ethanol	1.4	78.37	220	30.00	Oil	Pale green
5	Ether Petroleum	1.0	40-60	345	30.00	Gummy	Straw yellow
6	Ethyl acetate	1.2	76-77	240	30.00	Crude oil	Dark green
7	Hexane	1.2	65-70	230	30.00	Clear oil	Yellow
8	Methanol	1.1	64-66	200	30.00	Oil	Dark green
9	Aqua.*					Semi solid	Dark brown

Table 2 Contact toxicity of different solvent crude extracts of *Lantana camara* against the second instars of diamondback moth, *Plutella xylostella*

Solvents used for extraction	χ^2 at $p \leq 0.05$ (n=30)	Regression equation	LC ₅₀ %	95 per cent fiducial limit		LC ₉₅ %	95 per cent fiducial limit	
				LL	UL		LL	UL
Acetone	4.060	Y =1.422x+3.041	23.43	9.58	57.35	513.33	48.29	5456.45
Benzene	1.597	Y =1.353x+2.880	40.07	11.72	136.98	1496.1	64.35	34786.4
Chloroform	0.656	Y =1.631x+4.075	3.665	2.71	4.94	43.045	16.86	109.87
Ethanol	0.511	Y =1.413x+2.952	32.13	10.14	101.80	989.28	50.30	19454.3
Ethyl acetate	0.222	Y =1.686x+3.398	8.869	6.04	13.01	95.770	28.92	317.08
Hexane	1.514	Y =1.072x+4.597	1.567	0.55	4.45	266.56	8.85	8020.62
Methanol	3.619	Y =1.405x+3.744	7.673	5.24	11.22	103.78	27.09	397.58
Petroleum ether	0.330	Y =1.251x+3.240	31.44	8.96	110.22	1356.0	42.64	43116.9

Being compounds of natural origin, no problems with persistence in the environment is anticipated (Gebbinck *et al.*, 2002). Thus, products based on plant extracts, phyto-oils and purified substances of plant origin can be an alternative to the conventional pesticides (Isman, 2001). The crude plant extract consists of complex mixtures of active compounds. The complex mixtures act synergistically (Berenbaum, 1985) and show greater overall bioactivity compared to the individual components (Chen *et al.*, 1995). Also, there is less preference for insect to develop resistance against such mixtures (Shukla and Toke, 2013).

Ovicidal toxicity and oviposition deterrent activity

The studies reveal that the aqueous extract of *L. camara* leaf at 10 per cent concentration gave minimum egg hatch of 73.20 per cent, whereas at one per cent concentration, the egg hatch was maximum (81.20) and was at par with control (90.20) (Figure 3). However the hexane extract of this plant resulted in 15.33 per cent egg hatch at 10 per cent concentration, whereas it was 62.54 per cent at 1 per cent and 93.23 per cent in the untreated control. The neem oil resulted in 15.55 and 86.66 per cent egg hatch, respectively at 10 and 1 per cent concentrations in comparison to 91.11 per cent egg hatch in control, whereas the petroleum ether extract of this plant gave 13.33 and 77.50 per cent egg hatch at the respective concentrations in comparison to 90.83 per cent in control.

Among different solvent plant extracts of *L. camara* hexane extract was found to be more effective in detergency recording 2.33 and 15.00 egg/female/day at one and ten per cent respectively when compared to control (54.00 eggs). This was followed by ethyl acetate where mean no. eggs laid in leaf extract 1 and 10 per cent of *L. camara* was 0.00 and 20.67 respectively. When the comparison of aqueous and hexane extract was made, the hexane extract was found to be effective next to the neem and was significantly different with the other extract (Figure 4). An earlier study the used different solvents for preparation of test material against storage insect pest, *Cadra cautella* (Walker) of

wheat; the seed protection activity of *L. camara* extract from hexane was best reported by (Gotyal *et al.*, 2010) is supported for present study.

Azadirachtin, active neem constituent, has been reported to interfere with ecdysis of insects (Singh and Bhathal, 1994) and moulting disruption due to neem constituents has been observed in *Spodoptera frugiperda*, *Pectinophora gossypiella*, *Heliothis virescens* and *H. zea* (Kubo and Klocke, 1982), *S. aexampta* (Tanzubil and McCaffery, 1990), *S. littoralis* (Martinez and Van Emden, 2001). Elumalai *et al.* (2007) reported that oviposition deterrent activity of mentha and neem oils found to have more deterrent activity against the gravid moths of *S. litura* and their significance are apparent. It may be due to the consequence volatiles present in the oils which makes malfunctioning of the ovariole in female moths.

GC-MS Analysis

The phytochemical components present in the all crude extracts of *L. camara* in different solvents were identified by GC-MS. The results showed the presence of alkaloids, tannins, flavonoids, saponins, steroids and reducing sugar in the plant. Table 3 - 9 revealed the groups of secondary metabolites detected in each solvent fraction. The chromatogram (Figures 6 -12) and phytochemical components with their retention time, molecular weight and percentage of composition in different solvents extracts presented in (Tables 3 - 9). Six to nineteen major components elucidated viz., caryophyllene, caryophyllene oxide, selina-6-en-4-ol, 2-hexadecen-1-ol, hexatriacontane, tetrapentacontane, 1, 3-cyclohexadiene-1-carboxaldehyde, 6 S-2,3,8,8-tetramethyltricyclo[5.2.2.0(1,6)]undec-2-ene, benzene, 1,2-Benzenedicarboxylic acid, mono(2-ethylhexyl) ester, 2,6,10-Trimethyl,14-ethylene-14-pentadecne, 3-nonanone, phytol and squalene were identified in various crude extracts. Caryophyllene oxide has been reported as having analgesic, anti-inflammatory activity and antifungal activity against dermatophytes (Chavan *et al.*, 2010). It is also well known as a preservative in food, drugs and cosmetics (Yang *et al.*, 1999).

Conclusion

The use of persistent synthetic insecticides on vegetables and fruits is a concern due to practical limitations of the pre-harvest interval. Hence, the scientific communities working in the field of insect pest management have academic interest in the discovery and development of new bioinsecticides that are environmentally friendly to be integrated, in combination or rotation, with biopesticide segment. Experiment conducted to evaluate the insecticidal activity of *L. camara* leaves extracts against diamondback moth revealed high larval mortality, ovicidal effects and oviposition deterrence. Among the various solvents tested hexane crude extract showed maximum efficiency that was on par with neem oil.

The present study is the significant result of the extraction of *L. camara* leaves by soxhlet apparatus. The crude extracts are known to possess insect growth regulatory and strong oviposition deterency and further investigation have to be carryout to find the activity of crude form and to incorporate the IPM schedule. This trend is in line with the requirements of new regulations on Integrated Pest Management.

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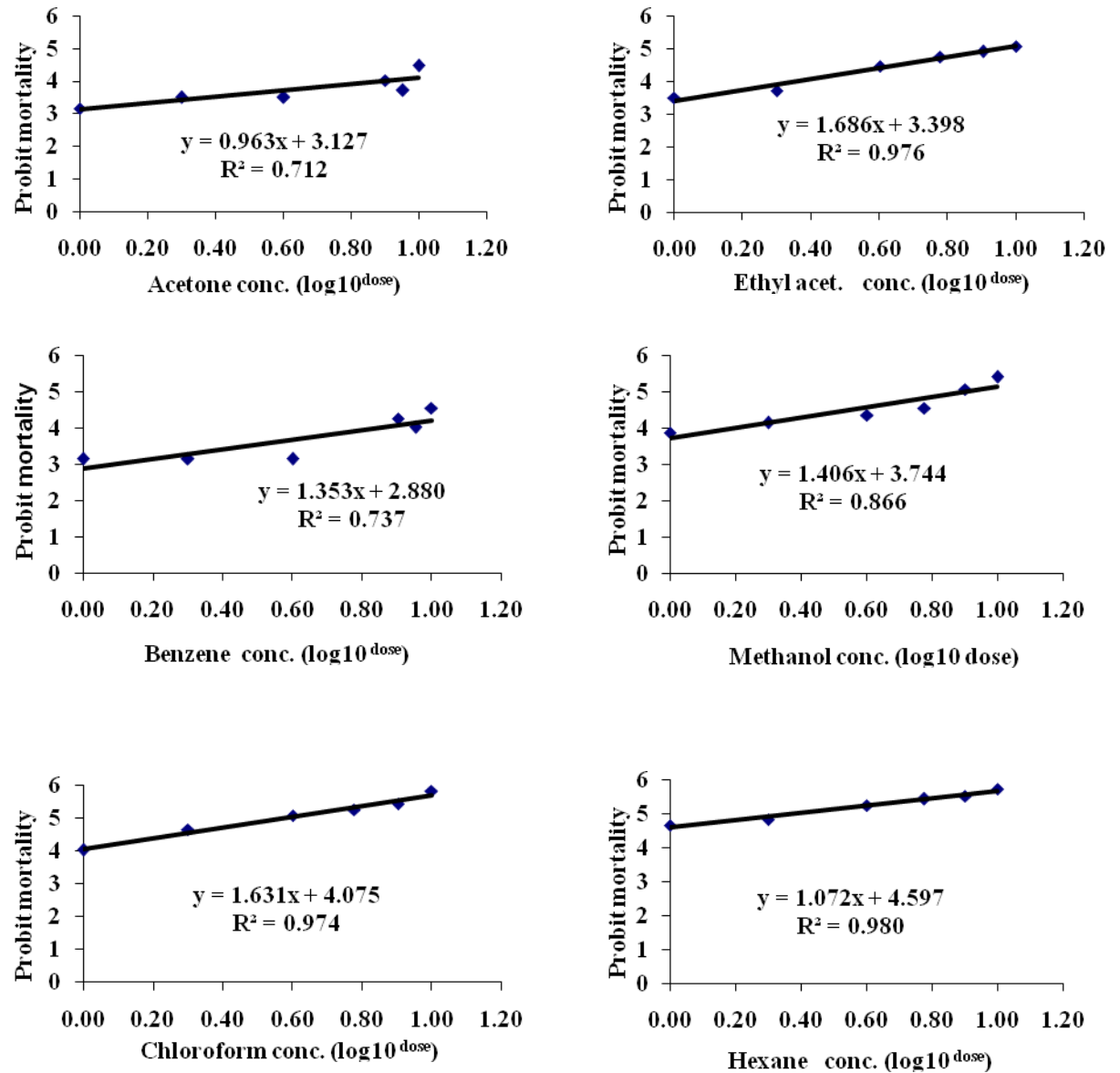
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Appendix

Figure 1. Concentration mortality response of *Lantana camara* L. crude extraction in different solvents against second instars of DBM



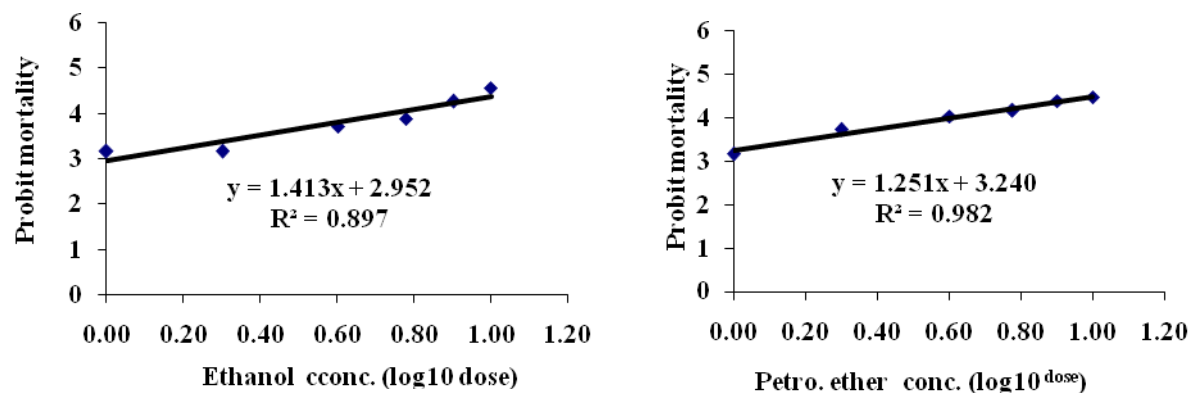
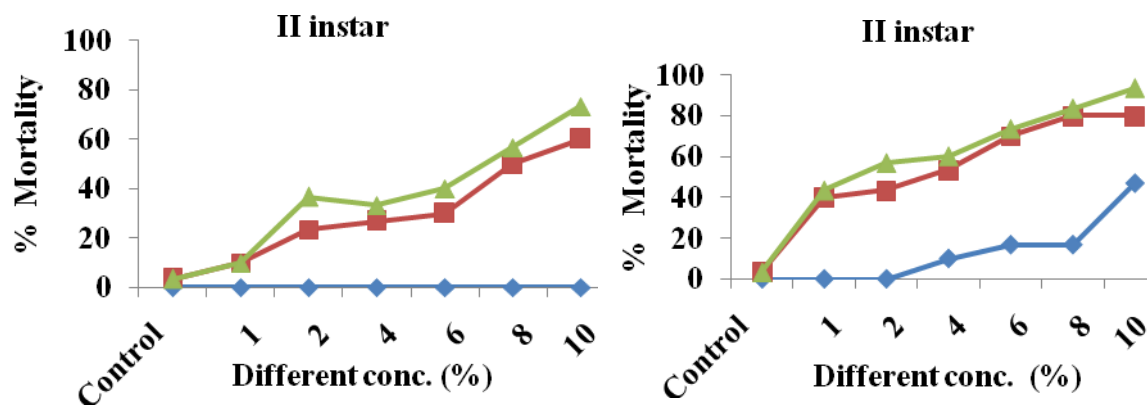


Figure 2. Toxicity of crude extracts of *Lantana camara* against diamondback moth, *Plutella xylostella*

a. Aqueous Extract

b. Acetone Extract



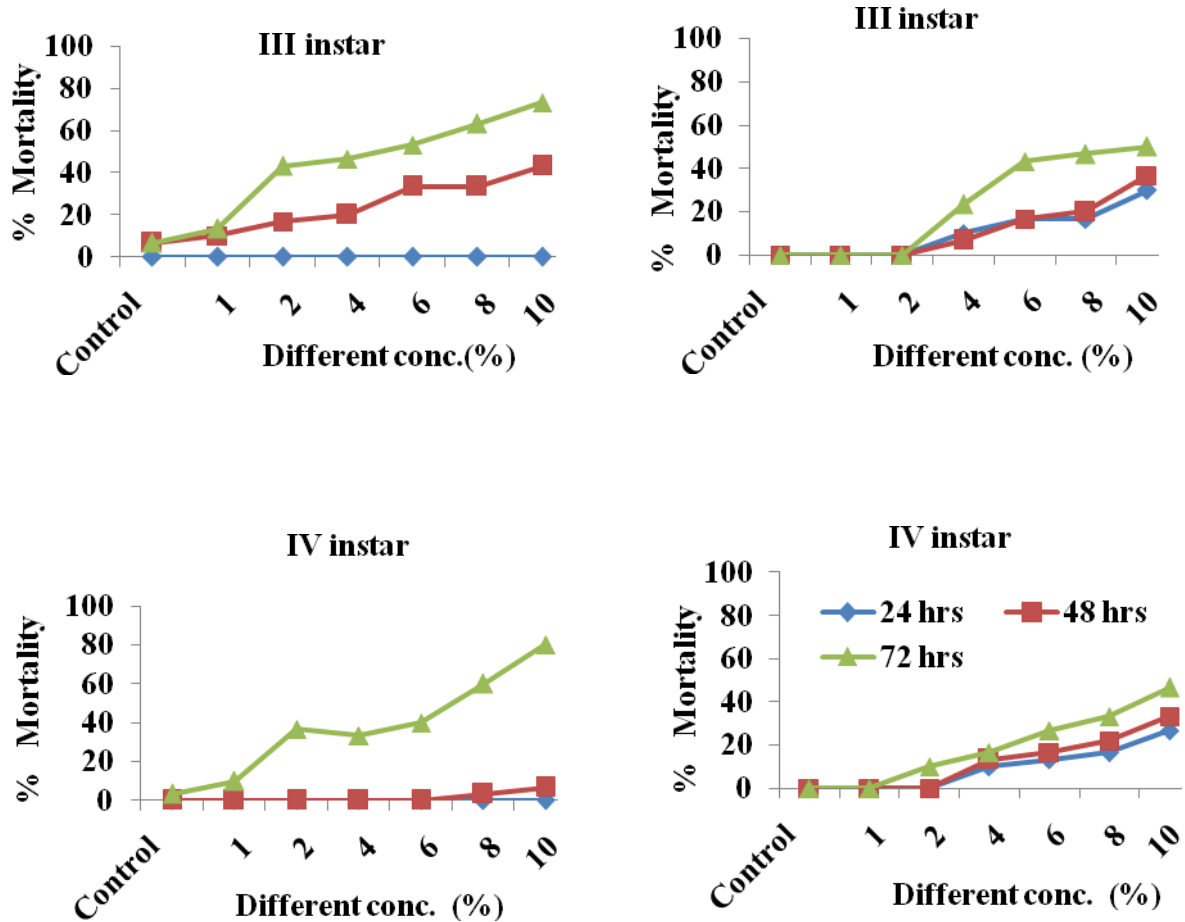
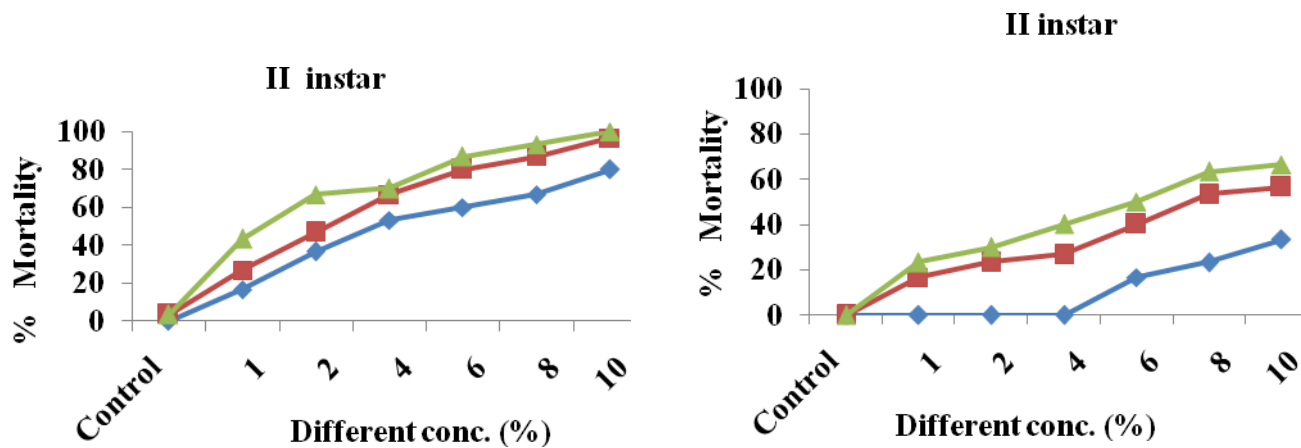


Figure 2. Toxicity of crude extracts of *Lantana camara* against diamondback moth, *Plutella xylostella*

c. Chloroform crude Extract

d. Benzene crude Extract



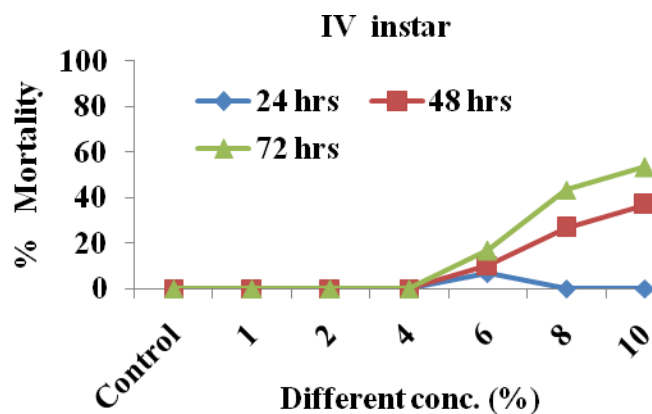
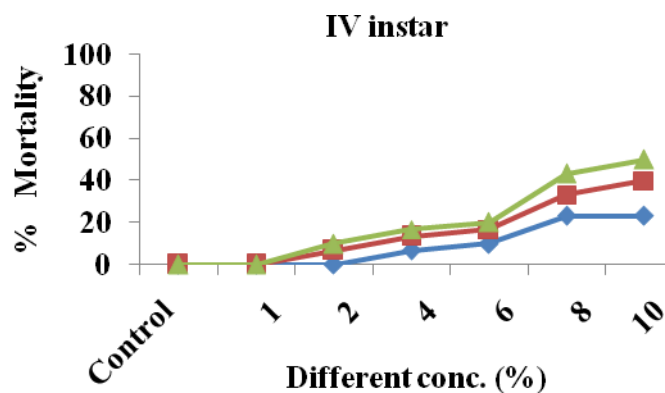
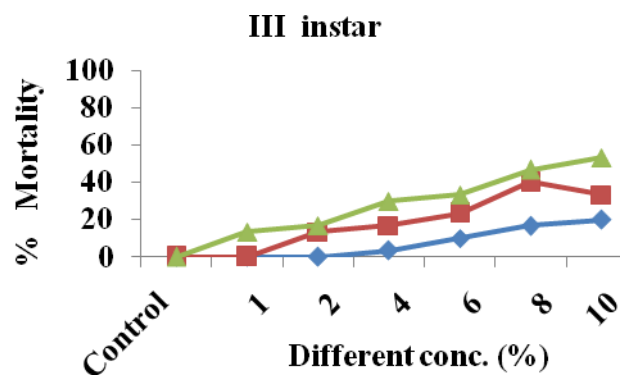
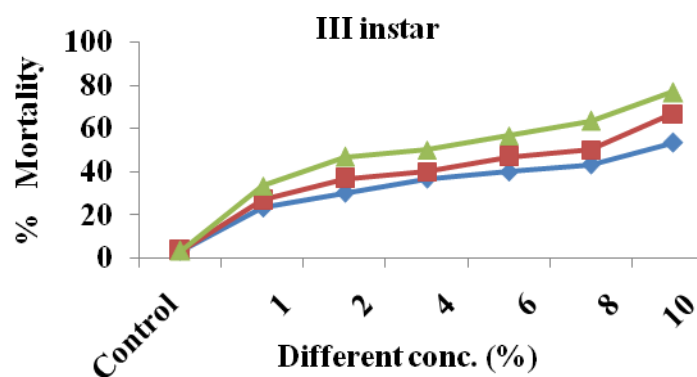


Figure 2. Toxicity of crude extracts of *Lantana camara* against diamondback moth, *Plutella xylostella*

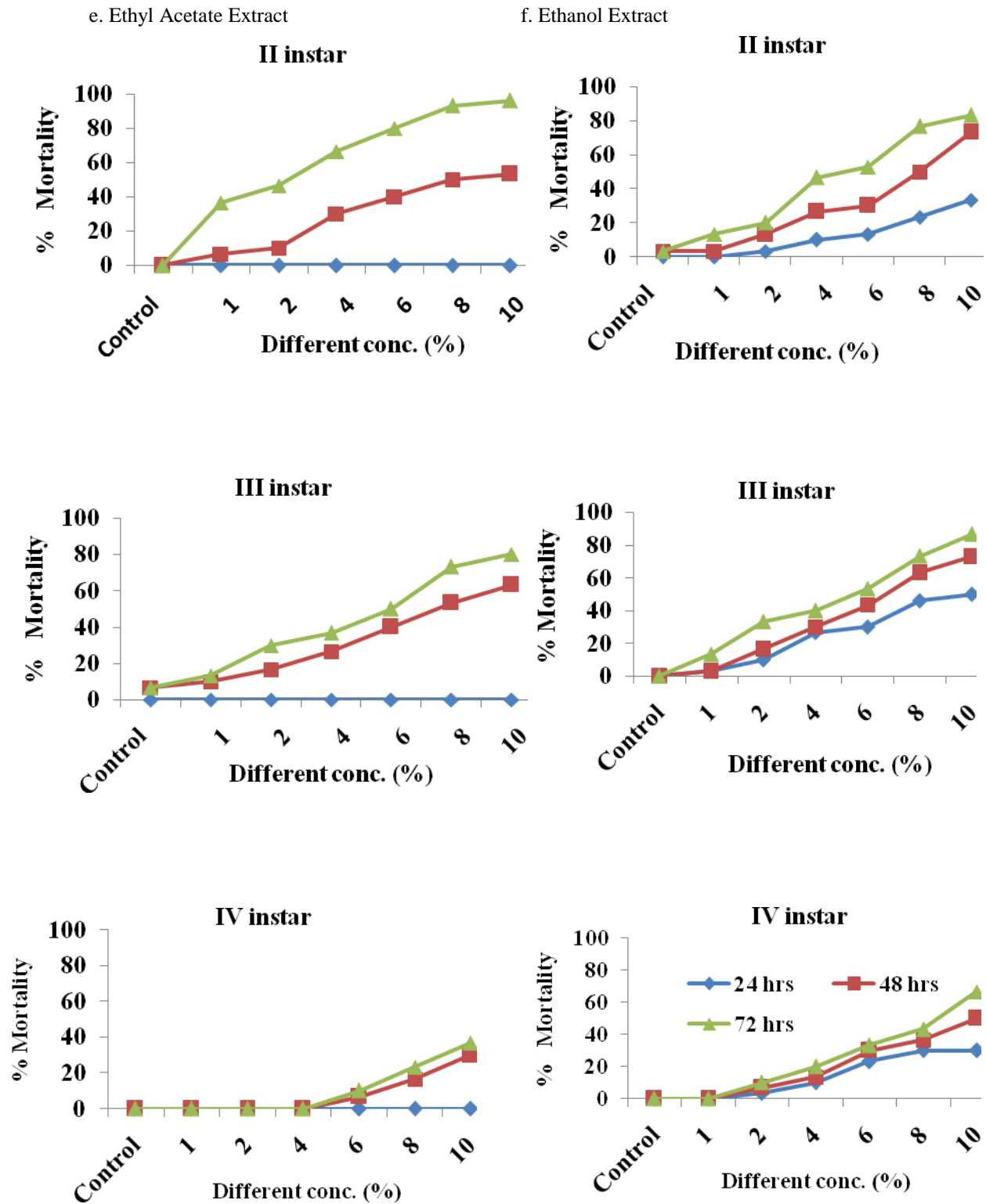


Figure 2. Toxicity of crude extracts of *Lantana camara* against diamondback moth, *Plutella xylostella*

g. Methanol Extract

h. Petroleum ether Extract

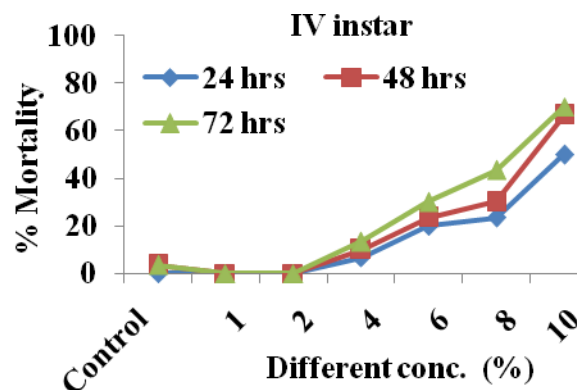
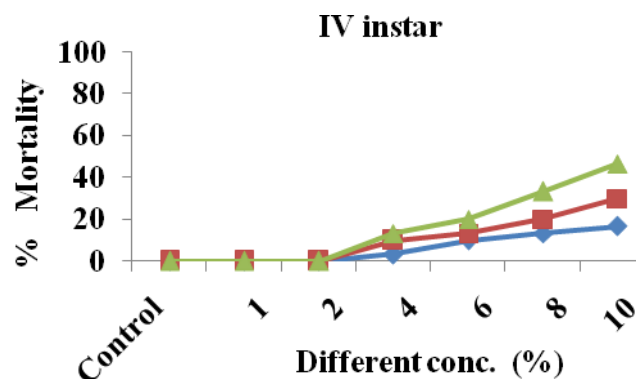
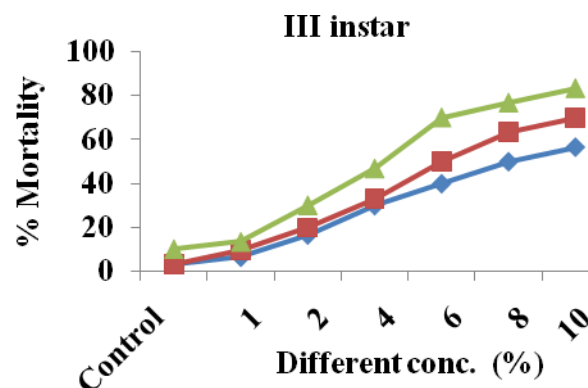
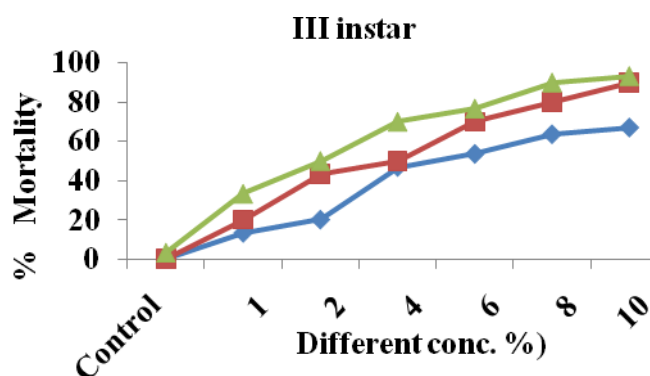
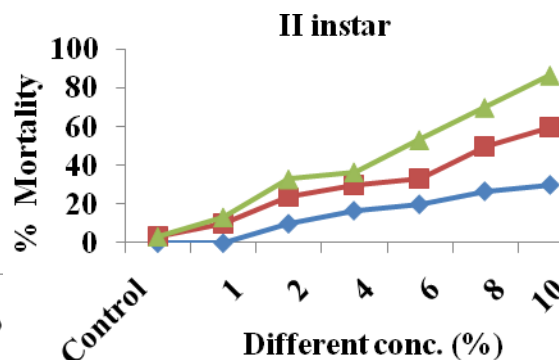
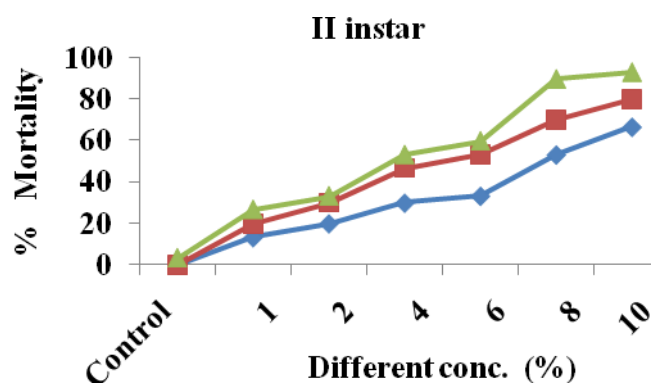


Figure 3. Insecticidal effects of different concentration with solvent crude extracts against the pupae of DBM, *Plutella xylostella* (L.)

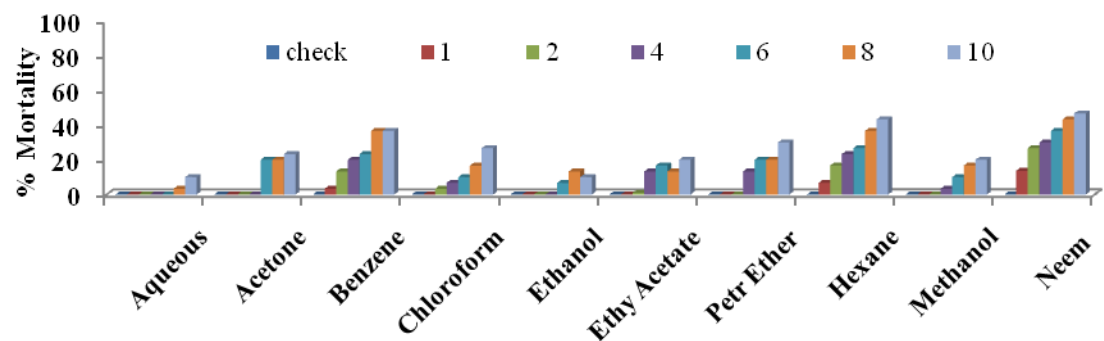


Figure 4. Ovicidal effects of different concentration with solvent crude extracts against the DBM, *Plutella xylostella* (L.)

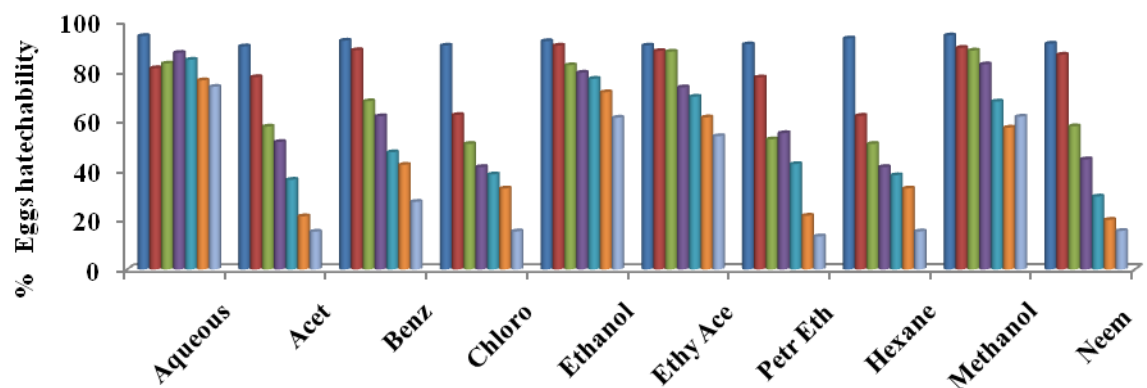


Figure 5. Oviposition deterency of different concentration with solvent crude extracts on 24hrs old adult DBM , *Plutella xylostella* (L.)

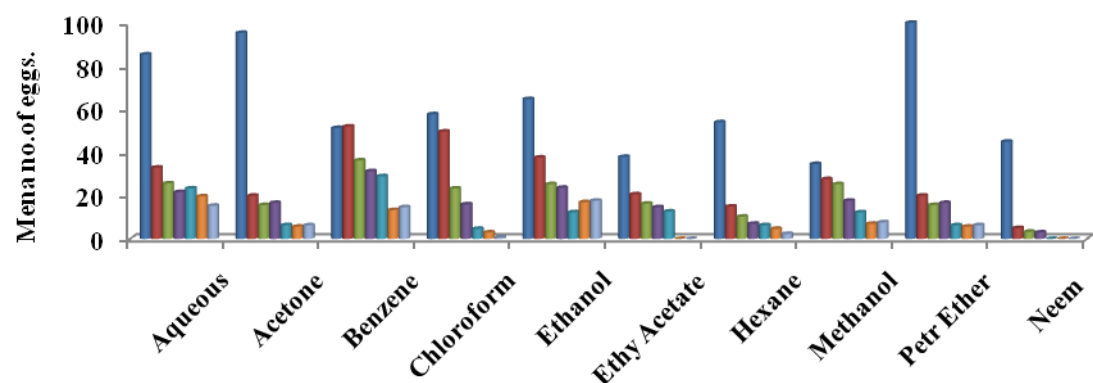


Table 3 Major phytochemical components identified in the acetone crude extract of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	5.412	69556	8.74	142	3- Nonanone
2	6.297	84859	10.67	98	3-Hexen-2-one
3	18.395	241744	30.39	204	Caryophyllene
4	28.437	118298	14.87	296	4-Hexen-1-ol
5	38.152	63813	8.02	268	Octadecane
6	39.288	217229	27.31	410	Squalene

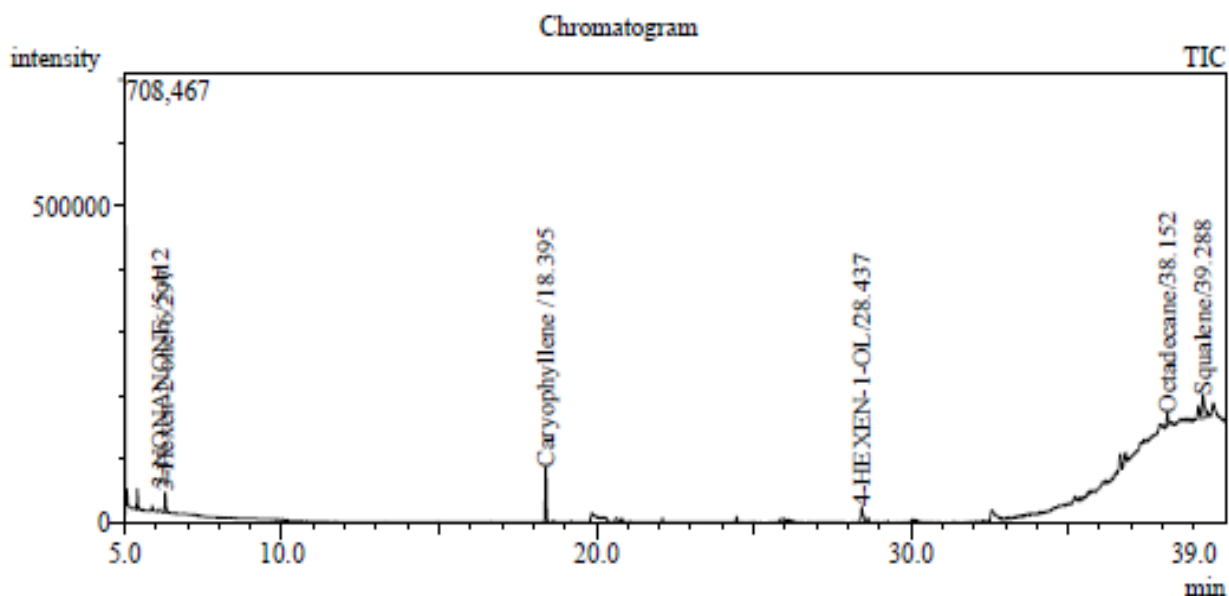


Figure 6. Gas chromatograph of *L. camara* acetone leaf extract

Table 4 Major phytochemical components identified in the benzene crude extract of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	5.413	129315	1.50	142	3- Nonanone
2	6.298	149176	1.73	98	3-Hexen-2-one
3	18.398	124393	1.44	204	Caryophyllene
4	33.168	1104850	12.78	408	6S-2,3,8,8-Tetramethyltricyclo[5.2.2.0(1,6)]undec-2-ene
5	34.860	1504455	17.41	404	Stigmast-5-en-3-ol
6	36.588	2874871	33.27	758	Tetrapentacontane
7	38.805	2755230	31.88	376	Bicyclo[4.1.0]Heptane

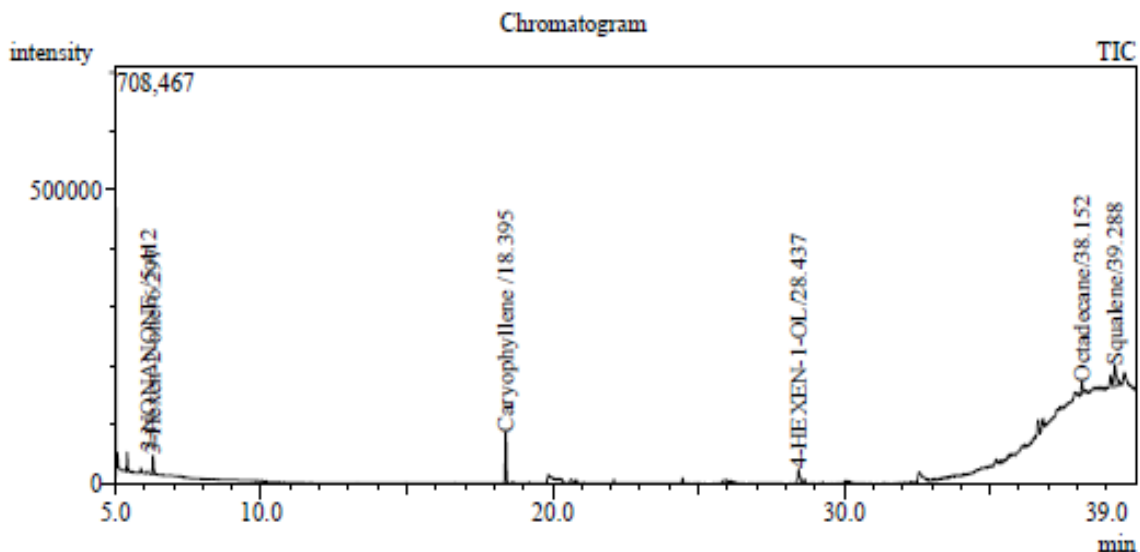


Figure 7. Gas chromatograph of *L. camara* benzene leaf extract

Table 5 Major Phytochemical components identified in the chloroform crude extract of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	5.411	84838	2.09	142	3- Nonanone
2	6.297	100721	2.48	98	3-Hexen-2-one
3	18.395	703969	17.34	204	Caryophyllene
4	18.636	34815	0.86	288	1,6-Cyclodecadiene
5	19.796	99508	2.45	202	1-(1,5-dimethyl-4-hexenyl)-4-methylbenzene
6	22.096	134177	3.31	220	(-)-5-Oxatricyclo [8.2.0.0(4,6)] dodecane
7	24.450	107785	2.66	190	3,7-Cyclodecadien-1-one
8	28.416	529755	13.05	278	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
9	29.249	104342	2.57	278	2,6,10-Trimethyl,14-ethylene-14-pentadecne
10	32.539	203174	5.00	296	Phytol
11	36.542	473621	11.67	278	1,2-Benzenedicarboxylic acid, mono(2-ethylhexyl) ester
12	37.579	1482762	36.53	758	Tetrapentacontane

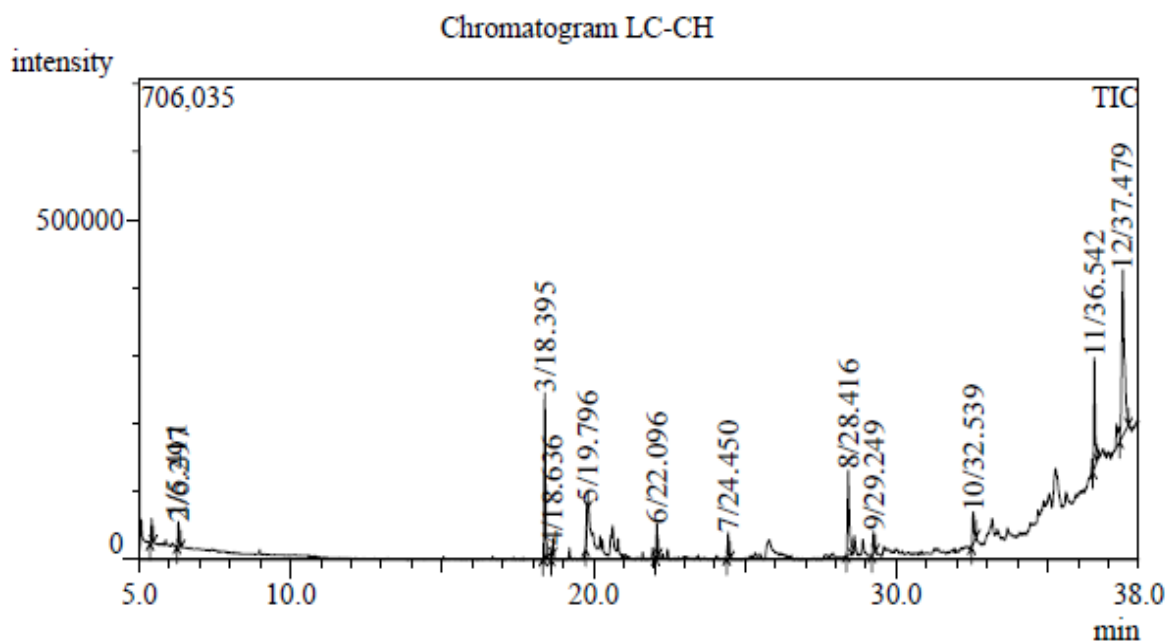


Figure 8. Gas chromatograph of *L. camara* chloroform leaf extract

Table 6 Major phytochemical components identified in the ethyl acetate crude extract of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	5.412	126557	2.47	116	3- Nonanone
2	6.298	141429	2.76	98	3-Hexen-2-one
3	18.400	224689	4.38	204	Caryophyllene
4	20.814	31406	0.61	218	A-Copaene
5	22.100	42748	0.83	220	2-Naphthaleneethanol
6	24.452	36104	0.70	150	1,3-Cyclohexadiene-1-carboxaldehyde
7	28.422	183526	3.58	278	2,6,10-Trimethyl,14-ethylene-14-pentadecne
8	32.544	106342	2.07	296	2-Hexadecen-1-ol
9	33.702	250132	4.88	278	2,6,10-Trimethyl,14-ethylene-14-pentadecne
10	35.235	1206182	23.53	758	Tetrapentacontane
11	36.547	340604	6.64	278	1,2-Benzenedicarboxylic acid, mono(2-ethylhexyl) ester
12	37.477	2436513	47.53	758	Tetrapentacontane

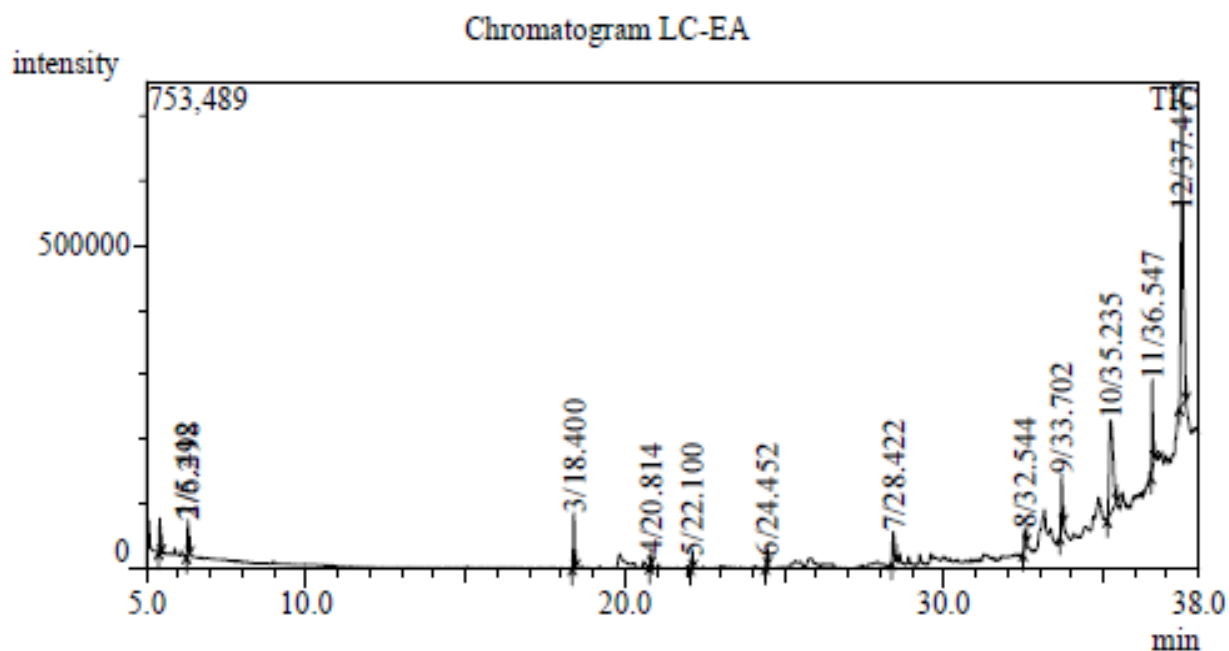


Figure 9. Gas chromatograph of *L. camara* ethyl acetate leaf extract

Table 7 Major phytochemical components identified in the hexane crude extract of *Lanatan camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	7.416	629223	3.72	142	Decane
2	8.026	289124	1.71	142	Octane
3	10.054	364146	2.15	170	Dodecane
4	18.395	2460437	14.54	204	Caryophyllene
5	19.776	2463832	14.56	202	Benzene,
6	20.561	577326	3.41	204	1,3-Cyclohexadiene
7	20.636	792782	4.69	272	1H-Benzocyclohepten-7-ol,
8	21.953	269683	1.59	220	1H-Cycloprop[e]azulen-7-ol,
9	22.096	596924	3.53	220	Caryophyllene oxide
10	22.275	301239	1.78	178	2-Cyclohexen-1-one,
11	22.447	268953	1.59	218	3,7-Cyclodecadien-1-one,
12	24.449	607914	3.59	190	3,7-Cyclodecadien-1-one,
13	25.706	2424352	14.33	204	1,5,5,9-Tetramethylspiro [5.5]undeca-1,8-dien
14	32.529	832065	4.92	296	Phytol
15	36.461	191438	1.13	338	Tetracosane
16	37.272	435561	2.57	506	Hexatriacontane
17	38.137	886886	5.24	506	Dotriacontane
18	39.117	1082388	6.40	450	Dotriacontane
19	39.271	1442021	8.52	450	2,6,10,14,18,22-Tetracosahexaene,

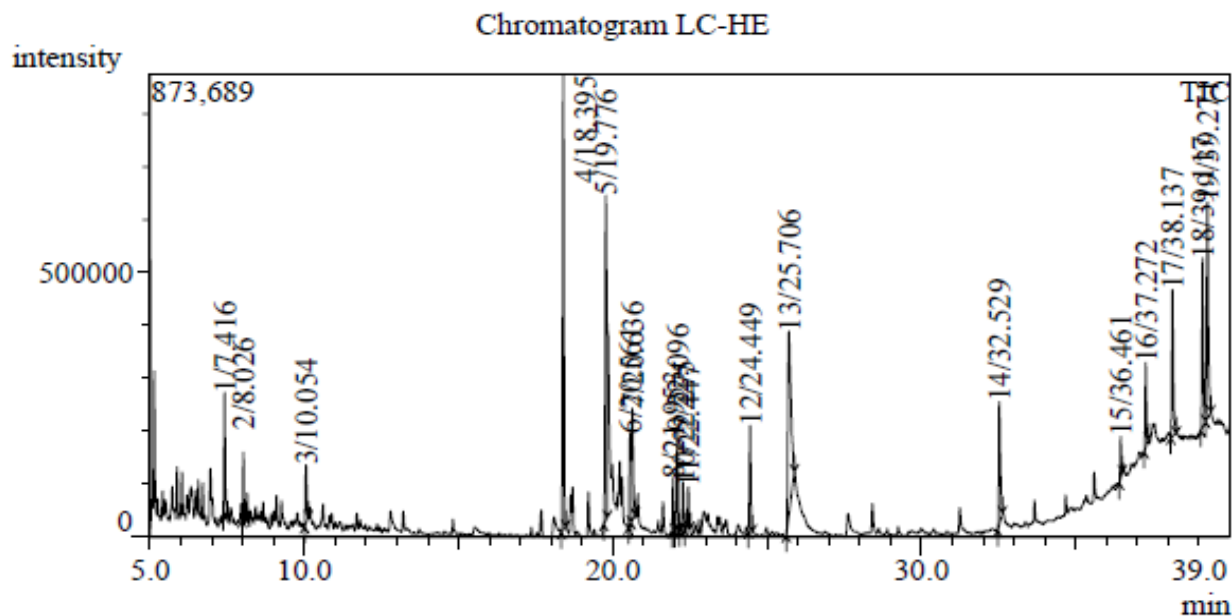


Figure 10. Gas chromatograph of *L. camara* hexane leaf extract

Table 8 Major Phytochemical components identified in the methanol crude extracts of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	5.408	128942	1.13	116	1-Pentanol,
2	6.293	156637	1.38	98	3-Hexen-2-one
3	18.391	867985	7.63	204	Caryophyllene
4	19.779	944108	8.30	208	Benzene
5	21.183	228871	2.01	222	Selina-6-en-4-ol
6	21.948	143428	1.26	220	1H-Cycloprop[e]azulen-7-ol,
7	22.091	363490	3.19	220	(-)-5-Oxatricyclo [8.2.0.0(4,6)] Dodecane
8	24.442	365690	3.21	190	3,7-Cyclodecadien-1-one
9	25.693	1702224	14.96	218	Phenol
10	28.410	254676	2.24	208	2,6,10-Trimethyl
11	32.523	488416	4.29	296	2-Hexadecen-1-ol
12	33.137	866945	7.62	204	6S-2,3,8,8-Tetramethyltricyclo [5.2.2.0(1,6)]undec-2-ene
13	34.852	1761406	15.48	376	Stigmast-5-en-3-ol, (3.beta.)
14	36.544	1236104	10.86	414	Tetrapentacontane

15 38.792 1870578 16.44 758 Cyclopentane-3'-spiropentacyclo

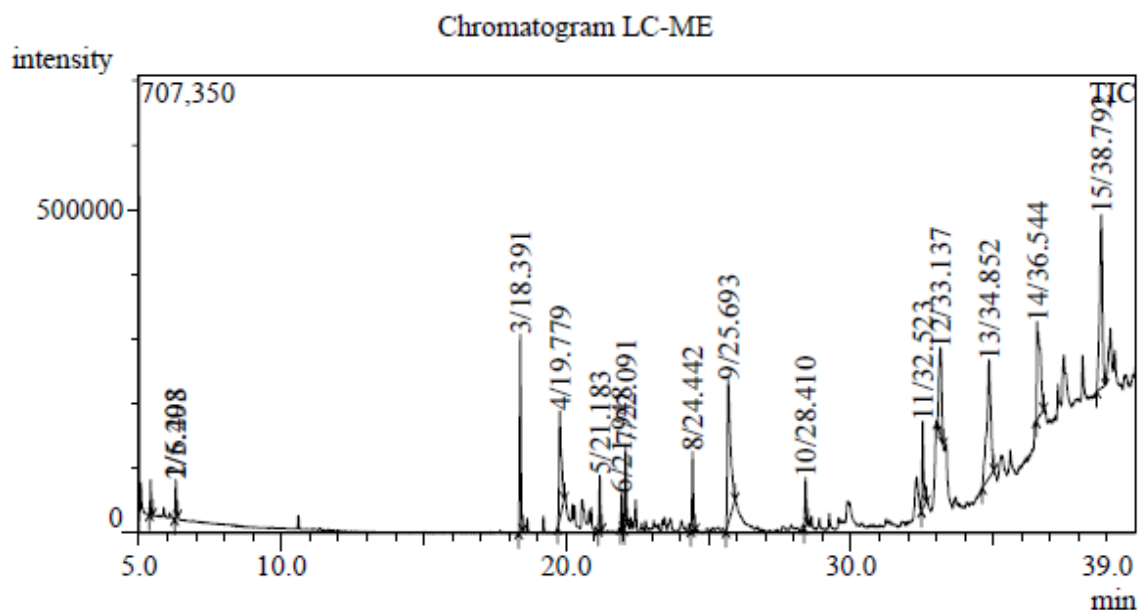


Figure 11. Gas chromatograph of *L. camara* methanol leaf extract

Table 9 Major Phytochemical components identified in the petroleum ether crude extract of *Lantana camara*

Peak no.	R. Time	Area	Area %	Molecular wt.	Components
1	18.396	1294144	15.99	204	Caryophyllene
2	19.790	747228	9.23	202	Benzene
3	19.850	475404	5.88	204	1,4-Methanoazulene
4	20.637	168648	2.08	204	1,4-Methanoazulene
5	22.097	296501	3.66	220	(-)-5-Oxatricyclo [8.2.0.0(4,6)] dodecane
6	24.451	258861	3.20	190	3,7-Cyclodecadien-1-one
7	25.720	1256391	15.53	296	Spiro[5.5]undeca-1,8-diene
8	32.532	328410	4.06	278	2-Hexadecen-1-ol,
9	36.542	1546546	19.11	506	1,2-Benzenedicarboxylic acid,
10	38.137	338233	4.18	410	Eicosane
11	38.786	450307	5.57	204	2-Cyclohexen-1-ol
12	39.120	409638	5.06	282	Hexatriacontane
13	39.271	521326	6.44	220	2,6,10,14,18,22-Tetracosahexaene,

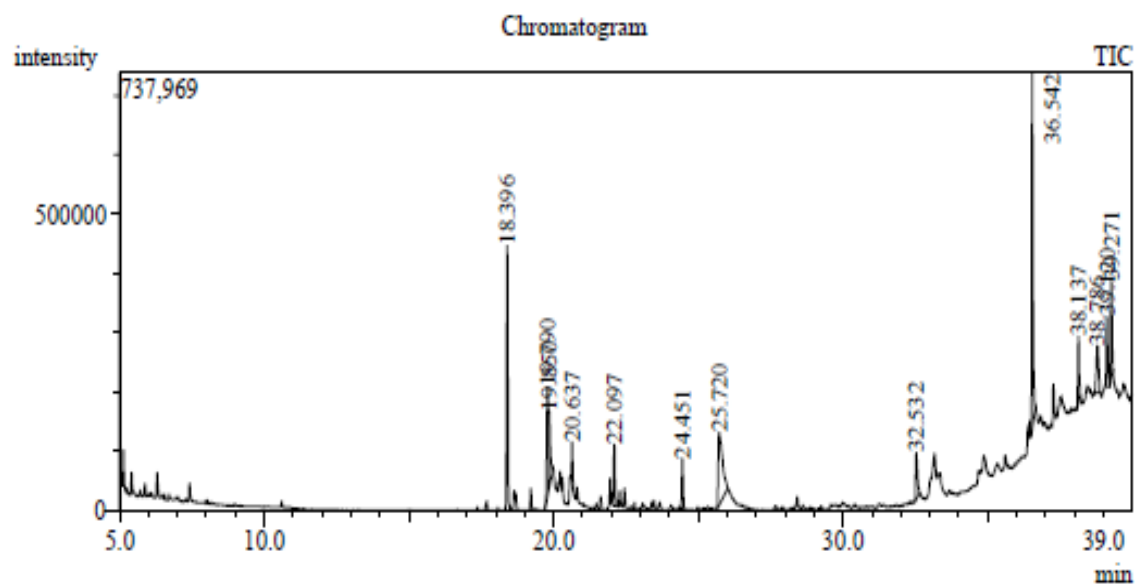


Figure 12. Gas chromatograph of *L. camara* petroleum ether leaf extract