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Biological activity of *Rosa arabica* (Rosaceae) Different Extracts against Rift Valley Fever Vector, *Culex antennatus* Becker (Diptera: Cuilicidae)

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ABSTRACT

Culex antennatus Becker is widely distributed in Egypt and significantly contributes to the incidence of RVF virus in the Nile Delta. The objective of this study was to assess the efficacy of leaf extracts of Rosa arabica (methanol, n-butyl acetate, chlorobenzene, and cyclohexane) against immature stages of C. antennatus. Additionally, the study aimed to determine the impact of the tested extracts on the reproductive potential of female *C. antennatus* offspring produced from larvae that were treated. The maximum larval mortality (100.0 percent) was observed at concentrations of 4500, 3000, 1800, and 1600 ppm of methanol, n-butyl acetate, chlorobenzene, and cyclohexane extract, respectively, according to the results obtained. The mean length period of both larvae and pupae was significantly ($P<0.05$) prolonged by all examined extracts in comparison to the untreated groups. Methanolic extract derived from the leaves of *R. arabica* demonstrated a statistically significant impact on female fecundity across all concentrations examined. Specifically, at 3500, 3000, and 2500 ppm, fecundity increased to 170.67±1.53, 178.50±1.29, and 180.00±2.39 eggs/♀, respectively, compared to 218.90±3.38 eggs/♀ for the control group. In addition, the hatchability of eggs was significantly improved by cyclohexane extract derived from the leaves of *R. arabica*, particularly at the two highest concentrations (1200 and 1400 ppm), where it was 82.10 and 80.86 percent, respectively, compared to 98.50 percent for the control. The cyclohexane extract exhibited the highest efficacy against *C. antennatus* immature stages and females derived from treated larvae, as indicated by the obtained results. The chlorobenzene, n-butyl acetate, and methanol extracts followed suit. In general, *R. arabica* extracts utilized in this investigation are regarded as novel and promising agents for suppressing *C. antennatus*, the mosquito vector.

INTRODUCTION

Egypt, together with other nations in the Eastern Mediterranean region as classified by the World Health Organization, is home to approximately 11.0 percent of all vector-borne diseases (WHO 2004). Numerous human and animal pathogenic agents, including dengue, malaria, filaria, Rift Valley Fever (RVF), and numerous arboviral illnesses, are transmitted by mosquitoes. These diseases account for hundreds of millions of clinical cases and millions of deaths annually (Dhanasekaran *et al.*, 2010; Roth *et al.*, 2010; Bakr *et al.*, 2014; Elhawary *et al.*, 2021; Shehata *et al.*, 2022). Moreover, the transfer of diseases by mosquitoes has

adverse effects on livestock, namely on the milk output of dairy cows and the reduction of weight increase (Islam *et al.*, 2017). *Culex antennatus* Becker (Diptera: Culicidae) is widely distributed in Egypt and significantly contributes to the prevalence of the RVF virus in the Nile Delta (Shehata *et al.*, 2019). *Culex antennatus* is also a principal vector of the Sindbis Virus and Western Nile Virus (WNV) (Harbach *et al.*, 1988; Fang *et al.*, 2022).

Historically, larvae of mosquito species are usually targeted using imitative chemical insecticides as a last resort to eliminate the rapid spread of diseases by vector control (Ravaomanarivo *et al.*, 2014). Nevertheless, the non-judicious application of chemical pesticides gives rise to a multitude of issues, including ecological contamination, insecticide resistance, and adverse health impacts on humans (Vijayakumar and Amirthanathn, 2014). Therefore, it is critical to identify alternative synthetic drugs that possess comparable bioactive properties but target distinct mosquito populations. Different plant extracts have several bioactivities and may act as larvicidal agents, fecundity suppression, repellent agents and enzyme inhibitory agents (Hassan *et al.*, 2014; Shehata 2018; Shehata *et al.*, 2020).

The objective of this study was to assess the efficacy of leaf extracts of *Rosa arabica* (Rosaceae), namely methanol, n-butyl acetate, chlorobenzene, and cyclohexane, against immature stages of *C. antennatus.* Additionally, the reproductive potential of *C. antennatus* females derived from larvae treated with the evaluated extracts was investigated.

MATERIALS AND METHODS

Culex antennatus Colonization:

The larvae of *Culex antennatus* were obtained from Faiyum Governorate, Egypt (Latitude: 29º18'53.4" N, Longitude: 30º39'19.2" E, Elevation: 19 m). Subsequently, the larvae were cultivated for five generations in the Medical Entomology Insectary, Animal House, Department of Zoology, Faculty of Science, Al-Azhar University. The conditions of the facility were strictly controlled, including temperature (27±2ºC), relative humidity (70±10 percent), and photoperiods (12:12). Following their emergence, adult mosquitoes were housed in wooden cages measuring 30 by 30 by 30 cm and were supplied daily with cotton pieces soaked in a solution containing 10.0 percent sucrose for a duration of 3 to 4 days. Females were thereafter permitted to have a blood meal from a pigeon host, which is essential for egg production (anautogeny). For egg-laying, a 20x20 cm plastic cup oviposition with dechlorinated tap water was positioned within the cage. After removing the egg rafts from the plastic plate, they were put into 25-by-30x15 centimeters plastic pans containing three liters of tap water and left for twenty-four hours. The larvae that were hatching were fed one piece of bread per day (Hassanain *et al.*, 2019).

Preparation of Tested Extracts:

The *Rosa arabica* specimen in this research was procured from Saint Catherine, South Sinai Governorate, Egypt, which is situated at an elevation of 1,586 and has a latitude of 28°33'42.88" N and a longitude of 33°56'57.62" E. A modified version of the conventional procedure outlined by Hassan et al. (2014) was to generate the extracts that were tested. The leaves of *R. arabica* were maintained at room temperature (26±2ºC) and shielded from sunlight for several days before being individually ground into powder using a hammer mill. Solvents like cyclohexane, chlorobenzene, methanol, and n-butyl acetate were in the extraction process. A quantity of one hundred grams of powdered leaves was extracted individually and subjected to five filtration cycles in 350 ml of room temperature solvent. The supernatants were decanted, filtered, and dried for 40-60 minutes, respectively, in a rotary evaporator set at 40ºC after 24 hours. The dry extracts were stored at -5ºC until they were utilised in the tests.

Biological Activity of Tested Extracts:

For determining the biological activity of the extracts under investigation, a conventional technique outlined by Elhawary *et al.,* (2021) was utilised. To facilitate the dissolution of the tested materials in water, two drops of Tween80 were used as an emulsifier to dissolve the compounds. Preparation of various quantities of each tested extract. Every substance that underwent testing was formulated in 500 ml plastic cups containing 250 ml of dechlorinated tap water. Twenty-five third-instar larvae of *C. antennatus* were subsequently transferred directly into plastic containers containing extracts of varying strengths. Triple repetitions of each test were conducted. Until the appearance of adults, daily mortality counts were collected for every plastic cup incubated under controlled conditions of a mosquito colony. 2 drops of $Tween_{80}$ were added to 250 ml of water for the control larvae. Mean±SD is used to calculate all values. A lack of response to mechanical stimulation or inability to mature into adults served as indicators of larval and pupal mortality. The mean value was then determined for the duration of each larva, which was determined by averaging the intervals between the onset of third instar larvae and the onset of pupation. The duration of pupal development was determined by calculating the time from the onset of pupation and the emergence of the adult. This number was then averaged for each individual.

Reproductive Potential of Resulted Females:

Females successfully hatching from *C. antennatus* third instar larvae treated with each concentration of the tested extracts were collected and, along with healthy males obtained from the colony, transferred to wooden cages measuring 20×20×20 cm. The cages were subsequently supplied with a 10.0 percent sugar solution for three days, after which both the males and females were left without sugar solution for one day, as recommended by the WHO. The starving females were permitted to have a blood meal from a pigeon and construct egg rafts using oviposition traps, which were clean water sources, within their cages on the fifth day. The number of eggs and rafts was determined using a binocular, and the means were then calculated (Shehata, 2018). The proportion of sterility was calculated as follows: According to Toppozada *et al.* (1966), the sterility percentage can be calculated as follows: a = number of eggs laid per female in the treatment group; b = percentage of hatched eggs in the treatment group; A = number of eggs laid per female in the control group; B = percentage of hatched eggs in the control group.

Statistical Analysis:

For the statistical analysis, all data were exposed to Graph Pad in Stat software, Inc. following the approach of Lentner *et al.,* (1982). The mean and standard deviation were used to represent the data. The data were assessed using ANOVA, as suggested by Bailey, (1981). SPSS V.22 was to encode and enter the data. For quantitative data, measures such as mean, median, standard deviation, and standard error were utilised, whilst frequency was employed to portray qualitative data. At $P < 0.05$, the criterion for statistical significance was established. LC_{50}, LC_{75}, and LC_{90} lethal concentrations were computed using multiple linear regressions (Finney, 1971).

RESULTS

Biological Activity of *Rosa arabica*-Tested Extracts:

The concentrations of methanol, n-butyl acetate, chlorobenzene, and cyclohexane extract at which the maximum larval mortality recorded (100.0 %) occurred at 4500, 3000, 1800, and 1600 ppm, respectively. The experimental groups that were not treated with any of the investigated extracts exhibited a substantially longer mean duration period of both larvae and pupae ($P<0.05$).

It was noted that the methanolic extract of *R. arabica* had a harmful impact on pupae produced by treated larvae; at 4000 ppm, the pupal mortality rate was the highest at 45.83 percent, and at 1000 ppm, it dropped to 5.74 percent. Furthermore, the methanolic extract of R. arabica inhibited the development of *Culex antennatus* larvae, pupae, and adults at all concentrations tested; the growth indices for these organisms were as follows: 5.73, 7.81, 10.57, 11.32, 11.95, 12.70, and 13.61 at concentrations of 4000, 3500, 3000, 2500, 2000, and 1000 ppm, respectively, compared to 16.04 for the control group. (Table 1).

Table 1: Effect of *Rosa arabica* methanol extract on some biological aspects of *Culex antennatus.*

Conc. (ppm)	Larval mort. (%)	Larval Duration	Pupal Mort. (%)	Pupal Duration	Adult Emergence (%)	Development Duration	Growth Index (a/b)
Control	0.0±0.0	4.27±0.15^{a}	0.0±0.0	1.98±0.14^{a}	100.0±0.0	6.24±0.29^{a}	16.04±0.75^{a}
1000	6.67±2.31	4.69±0.09^{b}	5.74±2.57	2.23±0.06^{a}	94.26±2.57	6.93±0.15^{c}	13.61±0.31^{c}
1500	12.0±4.0	4.82±0.07^{c}	10.69±3.12	2.33±0.06^{b}	89.31±3.12	7.15±0.01^{d}	12.70±0.05^{d}
2000	25.33±4.62	4.90±0.22^{d}	12.59±3.57	2.42±0.11^{c}	87.41±3.57	7.32±0.33^{d}	11.95±0.33^{d}
2500	36.0±4.0	5.22±0.20^{d}	16.84±4.44	2.50±0.16^{d}	83.16±4.44	7.72±0.05^{d}	11.32±0.08^{d}
3000	46.67±2.31	5.40±0.19^{d}	20.15±5.08	2.64±0.06^{d}	79.85±5.08	8.04±0.14^{d}	10.57±0.26^{d}
3500	61.33±11.55	5.63±0.08^{d}	32.69±8.33	2.68±0.08^{d}	67.31±8.33	8.32±0.15^{d}	7.81±1.21^{d}
4000	73.33±4.62	5.82±0.18^{d}	45.83±7.22	2.72±0.09^{d}	54.17±7.22	8.54±0.09^{d}	5.73±0.95^{d}
4500	100.0±0.0	---	---	---	---	---	---

Conc.: Concentration; ppm: particle per million; SD: standard deviation; mort.: mortality; a: non-significant (P>0.05); b: significant (P<0.05); c: highly significant (P<0.01); d: very highly significant (P<0.001). Means followed by the same letter in the same column are not statistically significant. All data is represented as Mean±SD.

As shown from the results, the highest pupal mortality percent recorded by *R. arabica* n-butyl acetate extract (53.33%) was induced at 2700 ppm, this percent decreased to 10.42% at 900 ppm. A retarded effect on the growth of larvae, pupae, and adult *C. antennatus* by *R. arabica* n-butyl acetate extract was observed especially at the highest concentrations (2700, 2400, and 1100 ppm), where the growth index recorded 5.32, 7.67, and 9.83 vs. 16.00 for the untreated group (Table 2).

Table 2: Effect of *Rosa arabica* n-butyl acetate extract on some biological aspects of *Culex antennatus.*

Conc. (ppm)	Larval mort. (%)	Larval Duration	Pupal Mort. (%)	Pupal Duration	Adult Emergence (%)	Development Duration	Growth Index (a/b)
Control	0.0±0.0	4.22±0.10^{a}	0.0±0.0	2.04±0.13^{a}	100.0±0.0	6.25±0.19^{a}	16.0±0.50^{a}
900	10.67±4.62	4.81±0.11^{d}	10.42±2.31	2.30±0.03^{a}	89.58±2.31	7.11±0.12^{c}	12.60±0.38^{d}
1200	18.67±8.33	4.91±0.10^{d}	11.76±4.26	2.37±0.07^{a}	88.24±4.26	7.28±0.17^{d}	12.17±0.56^{d}
1500	30.67±4.62	5.19±0.06^{d}	13.43±2.89	2.46±0.07^{b}	86.57±2.89	7.65±0.02^{d}	11.31±0.37^{d}
1800	40.0±4.0	5.32±0.17^{d}	17.98±4.80	2.54±0.15^{c}	82.02±4.80	7.86±0.32^{d}	10.54±0.45^{d}
2100	52.0±4.0	5.46±0.19^{d}	22.55±6.31	2.69±0.19^{d}	77.45±6.31	8.15±0.37^{d}	9.83±0.41^{d}
2400	68.0±4.0	5.69±0.11^{d}	37.90±4.78	2.79±0.16^{d}	62.10±4.78	8.48±0.14^{d}	7.67±0.82^{d}
2700	77.33±2.31	5.93±0.08^{d}	53.33±5.77	2.84±0.09^{d}	46.67±5.77	8.78±0.16^{d}	5.32±0.71^{d}
3000	100.0±0.0	---	---	---	---	---	---

See footnote of table (1).

There was a remarkable reduction in the adult emergence percent among the adults developed from larvae treated with *R. arabica* chlorobenzene extract at all concentrations as it recorded 35.0, 46.67, 75.35, 79.21, 82.92, 87.60, and 89.03 % at 600, 1400, 1200, 1000, 800, 600, and 400 ppm, respectively compared with 100.0% for the control congers (Table 3).

Table 3: Effect of *Rosa arabica* chlorobenzene extract on some biological aspects of *Culex antennatus.*

Conc. (ppm)	Larval mort. (%)	Larval Duration	Pupal Mort. (%)	Pupal Duration	Adult Emergence (%)	Development Duration	Growth Index (a/b)
Control	0.0±0.0	4.27±0.10^{a}	0.00±0.00	2.18±0.06^{a}	100.0±0.0	6.45±0.13^{a}	15.51±0.32^{a}
400	14.67±2.31	4.92±0.20^{d}	10.97±2.89	2.40±0.15^{a}	89.03±2.89	7.32±0.34^{d}	12.19±0.76^{d}
600	24.0±4.0	5.12±0.16^{d}	12.40±3.71	2.53±0.05^{b}	87.60±3.71	7.65±0.11^{d}	11.30±0.35^{d}
800	37.33±2.31	5.27±0.06^{d}	17.08±4.02	2.59±0.06^{c}	82.92±4.02	7.86±0.12^{d}	10.28±0.22^{d}
1000	48.0±4.0	5.39±0.06^{d}	20.79±5.72	2.69±0.13^{d}	79.21±5.72	8.08±0.09^{d}	9.81±0.82^{d}
1200	56.0±4.0	5.55±0.07^{d}	24.65±7.04	2.84±0.07^{d}	75.35±7.04	8.39±0.13^{d}	8.66±0.43^{d}
1400	77.33±2.31	5.78±0.17^{d}	53.33±5.77	2.90±0.05^{d}	46.67±5.77	8.68±0.13^{d}	6.00±1.47^{d}
1600	81.33±2.31	5.97±0.09^{d}	65.00±8.66	2.98±0.15^{d}	35.00±8.66	8.95±0.21^{d}	3.90±0.93^{d}
1800	100.0±0.0	---	---	---	---	---	---

See footnote of table (1).

On the other hand, the larval mortality recorded 85.33, 81.33, 64.00, 53.33, 41.33, 32.0, and 18.67 % at 1400, 1200, 1000, 800, 600, 400, and 200 ppm of *R. arabica* cyclohexane extract, respectively. The lethal effect of *R. arabica* cyclohexane extract was extended to the pupal stage, where the pupal mortality recorded 83.33, 65.00, 30.28, 22.98, 18.25, 13.87, and 11.51 % at 1400, 1200, 1000, 800, 600, 400, and 200 ppm, respectively. Also, mean pupal duration was prolonged by all concentrations used, where it recorded 3.10±0.10, 2.99±0.08, 2.94±0.05, 2.81±0.11, 2.77±0.15, 2.65±0.05, and 2.55±0.12 at 1400, 1200, 1000, 800, 600, 400, and 200 ppm, compared with 2.20±0.08 days for the control group (Table 4).

Table 4: Effect of *Rosa arabica* cyclohexane extract on some biological aspects of *Culex antennatus.*

Conc. (ppm)	Larval mort. (%)	Larval Duration	Pupal Mort. (%)	Pupal Duration	Adult Emergence (%)	Development Duration	Growth Index (a/b)
Control	0.0±0.0	4.35±0.11^{a}	0.0±0.0	2.20±0.08^{a}	100.0±0.0	6.55±0.16^{a}	15.27±0.36^{a}
200	18.67±2.31	5.08±0.07^{d}	11.51±3.03	2.55±0.12^{c}	88.49±3.03	7.63±0.06^{d}	11.60±0.47^{d}
400	32.0±4.0	5.24±0.05^{d}	13.87±4.24	2.65±0.05^{d}	86.13±4.24	7.89±0.09^{d}	10.86±0.44^{d}
600	41.33±2.31	5.40±0.15^{d}	18.25±4.32	2.77±0.15^{d}	81.75±4.32	8.17±0.29^{d}	10.03±0.10^{d}
800	53.33±2.31	5.54±0.05^{d}	22.98±5.58	2.81±0.11^{d}	77.02±5.58	8.35±0.08^{d}	9.23±0.74^{d}
1000	64.00±4.0	5.63±0.06^{d}	30.28±9.14	2.94±0.05^{d}	69.72±9.14	8.57±0.08^{d}	7.75±0.41^{d}
1200	81.33±2.31	5.87±0.19^{d}	65.00±8.66	2.99±0.08^{d}	35.0±8.66	8.85±0.22^{d}	4.89±0.66^{d}
1400	85.33±2.31	6.05±0.08^{d}	83.33±14.43	3.10±0.10^{d}	16.67±14.43	9.15±0.18^{d}	1.83±1.59^{d}
1600	100.0±0.0	---	---	---	---	---	---

See footnote of table (1).

From the results, it is obvious that the toxicity values of *R. arabica* tested methanol, n-butyl acetate, chlorobenzene, and cyclohexane extracts against *C. antennatus* larvae based on LC_{50}, LC_{75}, and LC_{90}values can be arranged in descending order as cyclohexane extract > chlorobenzene extract > n=butyl acetate extract >methanol extract (Table 5).

Table 5: Lethal concentrations (LC) of tested extracts from leaves of *Rosa arabica* against *Culex antennatus* third larval instar.

Extracts	LC_{50} (LCL-UCL)	LC_{75} (LCL-UCL)	LC_{90} (LCL-UCL)
Methanol	2938.81 (2663.11-3214.51)	3913.15 (3673.48-4152.81)	4497.75 (4276.38-4719.13)
N-Butyl Acetate	1956.85 (2893.22-2952.95)	2558.57 (2524.50-2592.63)	2952.95 (2893.22-2952.95)
Chlorobenzene	1019.16 (959.48-1078.84)	1434.72 (1417.22-1452.22)	1078.84 (1671.05-1697.04)
Cyclohexane	734.86 (693.13-776.58)	1169.47 (1136.50-1202.43)	1430.23 (1398.91-1461.54)

LCL: 95% Lower Confidence Limits; UCL: 95% Upper Confidence Limits. All values are represented as ppm, part per million.

Reproductive Potential of Resulted Females:

Methanolic extract from leaves of *R. arabica* exhibited a significant effect on female fecundity at all concentrations used, where the fecundity was 170.67±1.53, 178.50±1.29 and 180.00±2.39 eggs/♀ at 3500, 3000 and 2500 ppm, respectively vs. 218.90±3.38 eggs/♀ for the control. A decrease in the average number of hatched eggs as induced by the methanol extract from leaves of *R. arabica* was recorded. Also, the percentage of hatched eggs was 98.81% for the control group, this percentage decreased to 85.50, 86.83 and 87.46 at 3500, 3000 and 2500 ppm, respectively. The percent of sterility index for females resulting from treated larvae was increased by increasing the concentration, where it recorded 24.23, 26.88, 28.47 and 32.54% at 2000, 2500, 3000 and 3500 ppm, respectively (Table 6).

Table 6: Effect of methanol extract from leaves of *Rosa arabica* on the reproductive potential of *Culex antennatus* resulted from female.

Con. (ppm)	**No. of tested females**	**Eggs laid**		**Hatched eggs**			**Sterility Index (SI) %**
		Total	**Mean±SD**	**Total**	**Mean±SD**	**%**	
Control	10	2189	218.90±3.38^{a}	2163	216.30±3.71^{a}	98.81±0.33a	0.0
1000	8	1656	207.00±3.55^{b}	1552	194.00±3.55^{d}	93.72±1.04^{c}	10.31
1500	8	1530	191.25±5.26^{d}	1380	172.50±5.73^{d}	90.18±0.58^{d}	20.26
2000	7	1298	185.43±5.09^{d}	1147	163.86±3.53^{d}	88.38±1.15^{d}	24.23
2500	6	1085	180.0±2.39^{d}	949	158.17±3.31^{d}	87.46±0.81^{d}	26.88
3000	4	714	178.50±1.29^{d}	619	155.0±3.46^{d}	86.83±1.41^{d}	28.47
3500	3	512	170.67±1.53^{d}	444	146.0±3.61^{d}	85.50±2.25^{d}	32.54

See footnote of table (2).

Also, there was a significant ($P<0.01$) decrease in the mean number of eggs laid by females resulting from larvae treated with *R. arabica* n-butyl acetate extract at 1200, 1500, 1800, 2100 and 2400 ppm, where the average number was 188.88±0.83, 182.71±1.38, 177.60±2.07, 174.80±2.05 and 168.75±1.71 eggs/♀, respectively vs. 214.20±4.05 eggs/♀ for the control. There was a reduction in the hatchability percent as induced by n-butyl acetate extract from leaves of *R. arabica* was recorded, where it was 86.71, 85.47 and 84.30% at 1800, 2100 and 2400 ppm, respectively vs. 99.02% for the control (Table 7).

Table 7: Effect of n-butyl acetate extract from leaves of *Rosa arabica* on the reproductive potential of *Culex antennatus* resulted from female.

Con. (ppm)	No. of tested females	Eggs laid		Hatched eggs			Sterility Index (SI) %
		Total	Mean±SD	Total	Mean±SD	%	
Control	10	2142	214.20±4.05[a]	2122	212.10±3.57[a]	99.02±0.33[a]	0.0
900	10	2049	204.90±2.23[c]	1907	190.70±3.65[a]	93.06±1.06[d]	10.10
1200	8	1511	188.88±0.83[d]	1347	168.38±2.45[d]	89.15±1.20[d]	20.62
1500	7	1279	182.71±1.38[d]	1123	160.43±2.82[d]	87.80±1.08[d]	24.37
1800	5	888	177.60±2.07[d]	770	154.00±3.00[d]	86.71±1.36[d]	27.40
2100	5	874	174.80±2.05[d]	747	149.40±2.97[d]	85.47±1.41[d]	29.56
2400	4	675	168.75±1.71[d]	569	142.25±0.50[d]	84.30±0.87[d]	32.93

See footnote of table (2).

A significant (P<0.001) reduction in the number of eggs laid by the females emerged from larvae treated with chlorobenzene extract from leaves of *R. arabica* was recorded, the average number was 166.50±2.08, 170.75±1.26 and 175.17±2.71 eggs/♀ at the highest concentrations (1400, 1200 and 1000 ppm) compared with 211.20±1.62 eggs/♀ for the untreated group. Also, an increase in the percentage of sterility index was observed, where it was 26.70, 29.67 and 32.35% at 1000, 1200 and 1400 ppm, respectively (Table 8).

Table 8: Effect of chlorobenzene extract from leaves of *Rosa arabica* on the reproductive potential of *Culex antennatus* resulted female.

Con. (ppm)	No. of tested females	Eggs laid		Hatched eggs			Sterility Index (SI) %
		Total	Mean±SD	Total	Mean±SD	%	
Control	10	2112	211.20±1.62[a]	2051	205.10±1.66[a]	97.11±0.46[a]	0.0
400	9	1820	202.22±1.30[d]	1683	187.00±3.39[d]	92.47±1.39[c]	8.83
600	7	1302	186.00±0.82[d]	1148	164.00±1.41[d]	88.17±0.99[d]	20.04
800	9	1629	180.89±1.62[d]	1419	157.67±1.00[d]	87.16±0.37[d]	23.13
1000	6	1051	175.17±2.71[d]	902	150.33±3.44[d]	85.82±1.43[d]	26.70
1200	4	683	170.75±1.26[d]	577	144.25±0.96[d]	84.48±0.93[d]	29.67
1400	4	666	166.50±2.08[d]	555	138.75±2.50[d]	83.34±1.28[d]	32.35

See footnote of table (2).

In addition, a high effect of cyclohexane extract from leaves of *R. arabica* on egg hatchability especially at the two highest concentrations (1200 and 1400 ppm), where it was 82.10 and 80.86% vs. 98.50% for the control. A marked increase in the percentage of sterility index for females resulted from treated larvae especially at the two highest concentrations (1200 and 1400 ppm) were observed, where the sterility % was 35.62 and 37.65, respectively (Table 9).

Table 9: Effect of cyclohexane extract from leaves of *Rosa arabica* on the reproductive potential of *Culex antennatus* resulted from female.

Con. (ppm)	No. of tested females	Eggs laid		Hatched eggs			Sterility Index (SI) %
		Total	Mean±SD	Total	Mean±SD	%	
Control	10	2133	213.30±1.64[a]	2101	210.10±1.52[a]	98.50±0.29[a]	0.0
200	10	1997	199.70±1.25[d]	1826	182.60±3.24[d]	91.43±1.16[d]	13.12
400	10	1827	182.70±2.71[c]	1590	159.00±3.06[d]	87.02±0.64[d]	24.33
600	8	1417	177.13±1.73[d]	1218	152.25±2.12[d]	85.95±0.60[d]	27.54
800	6	1032	172.0±1.41[d]	873	145.50±1.38[d]	84.59±0.74[d]	30.75
1000	6	1004	167.33±1.21[d]	835	139.17±2.48[d]	83.17±1.33[d]	33.76
1200	4	659	164.75±0.96[d]	540	135.25±0.50[d]	82.10±0.69[d]	35.62
1400	2	324	162.0±0.0[d]	262	131.00±0.0[d]	80.86±0.0[d]	37.65

See footnote of table (2).

DISCUSSION

The findings demonstrated that all tested extracts evoked a promising larvicidal activity against *Culex antennatus* larvae. The toxicity of the tested extracts varied according to the solvent used in the extraction and the concentration of the extract. According to LC_{50}, LC_{75} andLC_{90}calculated values, cyclohexane extraction from leaves of *Rosa arabica* was more effective against *C. antennatus* third larval instar than those of chlorobenzene, n-butyl acetate, and methanol. The recorded effect of *R. arabica* extracts on *C. antennatus* third larval instar was harmonious with earlier results recorded by many authors using different plant extracts against several mosquito species. As an illustration, Al-Obaidi, (2019) evaluated the toxicity of *Silybum marianum* and *Nerium oleander* extracts against *C. quinquefasciatus* larvae and he found that *S. marianum* and *N. oleander* recorded LC_{50} of 57, 214, and 379 ppm for *S. marianum* against three instars, while LC_{90} values were 2422, 2936, and 3161 ppm against three instars, respectively after 24 h. On the other hand, *N. oleander* recorded LC_{50} values of 161, 194, and 360 ppm against three instars and LC_{90} values of 1968, 2004, and 4203 ppm, respectively; Hassanain *et al.,* (2019) used petroleum ether extract from leaves of *Lantana camara* against larvae of *Anopheles multicolor* and the highest larval mortality (100.0%) achieved by 140 ppm; Izah and Youkparigha, (2019) studied the larvicidal activity of fresh aqueous and ethanolic extracts of *Cymbopogon citratus* against *A. gambiae* and they observed that mortality rate at 50, 100, 150, 200 and 250 ppm was 23.33, 36.67, 48.33, 63.33 and 71.67%, respectively (aqueous extract), and 23.33, 50.0, 55.0, 76.67 and 85.0%, respectively (ethanolic extract). The ethanolic extract had superior activity compared to the aqueous extract with LC_{50} values of 104.47 and 161.06 ppm, respectively; Shehata (2019) recorded that petroleum ether extract from leaves of *Prunus domestica* and *Rhamnus cathartica* was more effective against *C. pipiens* (LC_{50} 33.3 and 63.4 ppm) than chloroform (LC_{50} 70.8 and 192.1 ppm) and methanolic extracts (LC_{50} 132.7 and 273.5 ppm); Shehata *et al.,* (2021) studied the insecticidal activity of *Pyrus communis* hexane (PCH) and methanol (PCM) extracts against *A. pharoensis* and they reported that tested PCH and PCM possess larvicidal activity against *An. pharoensis* with LC_{50} and LC_{90} values of 179.9, 41.2 and 314.1, 68.9 ppm, respectively, and Moola *et al.,* (2023) examined the chemical composition and larvicidal activity of *Hyptis suaveolens* essential oil against *Aedes aegypti* larvae and they found that the highest mortality rate was recorded at 100 ppm (LC_{50} of 32.85 ppm and LC_{90} of 66.54 ppm).

Also, treatment of *C. antennatus* third larval instar with tested extracts resulted in prolongation in both larval and pupal durations as compared with control congers; the prolongation in larval and pupal periods was solvent and concentration-dependent and was similar to that reported by Sharma *et al.,* (2006 a&b) using petroleum ether extract of *Artemisia annua* against *A. stephensi* and *C. quinquefasciatus* larvae, Coria *et al.,* (2008) using ethanolic extract of *Melia azedarach* leaves on *Ae. Aegypti* larvae, Coelho *et al.,* (2009) using *Moringa oleifera* lectin against *Ae. aegypti* larvae, Hassanain *et al.,* (2019) using petroleum ether extract from leaves of *L. camara* against larvae of *A. multicolor,* Shehata, (2019) using methanol, chloroform, and petroleum ether extracts from leaves of *Pr. domestica* and *Rh. cathartica* against *C. pipiens,* Shahat *et al.,* (2020) using *Otostegia fruticosa* leaves extracts against *C. pipiens* and Shahata *et al.,* (2021) using *Py. communis* PCH and PCM extracts against *A. pharoensis.* In addition, the toxic effect of tested extracts extended to *C. antennatus* pupae depending on the solvent used in the extraction and the concentration of the extract. Pupal mortality percentages recorded in the present study confirm those reported by Asiry *et al.,* (2017) using ethanolic leaf extracts of *Citrullus colocynthis, Artemisia annua, Pergularia tomentosa* and *Rhanterium epapposum* against *Ae. Aegypti,* Shehata, (2019) using methanol, chloroform, and petroleum ether extracts from

leaves of *Pr. domestica* and *Rh. cathartica* against *C. pipiens,* Shahat *et al.,* (2020) using *O. fruticosa* leaves extracts against *C. pipiens* and Shahata *et al.,* (2021) using *Py. communis* PCH and methanol PCM extracts against *An. pharoensis.*

On the other hand, all *R. arabica* extracts tested against 3rd instar larvae of *C. antennatus* significantly reduced the fecundity and increased the sterility % of females developed from treated larvae as compared with the untreated control. The fecundity and sterility percents were solvents used in extraction and were concentration-dependent. Moreover, a remarkable decrease in the hatchability % of eggs laid by females resulting from treated larvae with tested extracts used was observed. The hatchability % of eggs decreased as the concentration of the extract increased. These results are consistent with those obtained by many authors using different plant extracts against different mosquito species, for example, Coria *et al.,* (2008); Pavela, (2009); Shahat *et al.,* (2020) and Elhawary *et al.,* (2021).

Conclusion:

The extracts of *Rosa arabica* utilised in this research are regarded as novel and promising agents for suppressing the mosquito vector, *Culex antennatus.* Additionally, it is imperative that we devise and commence additional research endeavors in the near future that may yield bioactive chemicals in *R. arabica* extracts that may account for the observed biological activity, and then evaluate these compounds against a wide range of mosquito species.

Declarations:

Ethical Approval: The study followed the ethical customs of Faculty of Science, Al-Azhar University.

Competing interests: The authors declare no conflict of interest.

Authors Contributions: I hereby verify that all authors mentioned on the title page have made substantial contributions to the conception and design of the study, have thoroughly reviewed the manuscript, confirm the accuracy and authenticity of the data and its interpretation, and consent to its submission.

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EFERENCES

Al-Obaidi, M.J. (2019). Toxicity of *Silybum marianum* and *Nerium oleander* extracts against Mosquito Larvae of *Culex quinquefasciatus* (Say). *Proceedings of the Zoological Society,* 1294(6):62008. http://dx.doi.org/10.1088/1742-6596/1294/6/062008

Asiry, K.A., Hassan, S.S.M., Ibrahim, N.A., Al- Khuraiji, I.A., Kehial, M.A., Al-Anazi, N.A., Al-Nasser, A.S. and Al-Shehri, A.Z. (2017). Larvicidal efficacy of ethanolic leaf extracts of four selected local plants from hail region, northern Saudi Arabia, against the dengue fever vector, *Aedes aegypti* (L.) under laboratory conditions. *International Journal of Mosquito Research,* 4(3):81-87.

Bailey, R.A. (1981). A unified approach to design of experiments. *Journal of the Royal Statistical Society. Series A (General),* 144(2):214-223. https://www.jstor. org/ stable/i349598

Bakr, R.F.A., Nassar, M., El-Barkey., N.M., Kotb, T.F., Badrawy, H. and Abdeldayem, M.S. (2014). Prevalence of mosquitoes in Jazan Province, Saudi Arabia. *Egyptian Academic Journal of Biological Sciences A. Entomology,* 7(2):15-27. https:// doi.org/10.21608/eajbsa.2014.12940

Coria, C., Almiron, W., Valladares, G., Carpinella, C., Luduen˜a, F., Defago, M. and Palacios, S. (2008). Larvicide and oviposition deterrent effects of fruit and leaf extracts from *Melia azedarach* L. on *Aedes aegypti* (L.) (Diptera: Culicidae). *Bioresource Technology*, 99:3066-3070. https://doi.org/10.1016/j.biortech. 2007. 06.012

Dhanasekaran, D., Sakthi, V., Thajuddin, N. and Panneerselvam, A. (2010). Preliminary evaluation of *Anopheles* mosquito larvicidal efficacy of mangrove actinobacteria. *International Journal of Applied Biology and Pharmaceutical Technology*, 1(2):374-381.

Elhawary, E.A., Mostafa, N.M., Shehata, A.Z.I., Labib, R.M. and Singab, A.N.B. (2021). Comparative study of selected Rosa varieties' metabolites through UPLC-ESI-MS/MS, chemometrics and investigation of their insecticidal activity against *Culex pipiens* L. *Jordan Journal of Pharmaceutical Sciences,14*(4):417-433. https:// journals.ju.edu.jo/JJPS/article/view/107758

Fang, Y., Khater, E.I.M., Xue, J.B., Ghallab, E.H.S., Li, Y.Y., Jiang, T.G. and Li, S.Z. (2022). Epidemiology of Mosquito-Borne Viruses in Egypt: A Systematic Review. *Viruses*, 14:1577. https://doi.org/10.3390/v14071577

Finney, D.J. (1971). Probit Analysis. Third Edition. Cambridge University Press.

Harbach, R., Harrison, B., Gad, A., Kenawy, M. and El-Said, S. (1988). Records and notes on mosquitoes (Diptera: Culicidae) collected in Egypt. *Mosquito systematics*, 20: 317-342. https://apps.dtic.mil/sti/pdfs/ADA221832.pdf

Hassan, M.I., Fouda, M.A., Hammad, K.M., Tanani, M.A. and Shehata, A.Z. (2014). Repellent effect of *Lagenaria siceraria* extracts against *Culex pipiens*. *Journal of the Egyptian Society of Parasitology*, 44(1):243-248.https://doi.org/10. 21608/ jesp.2014.90754

Hassanain, N.A.E.H., Shehata, A.Z., Mokhtar, M.M., Shaapan, R.M., Hassanain, M.A.E.H. and Zaky, S. (2019). Comparison between insecticidal activity of *Lantana camara* extract and its synthesized nanoparticles against anopheline mosquitoes. *Pakistan Journal of Biological Sciences*, 22(7):327-334.https://dx.doi.org/10.3923/pjbs. 2019.327.334

Islam, J., Zaman, K., Duarah, S., Raju, P.S. and Chattopadhyay, P. (2017). Mosquito repellents: An insight into the chronological perspectives and novel discoveries. *Acta Tropica*, 167:216-230. https://doi.org/10.1016/j.actatropica. 2016. 12. 031

Izah, S.C. and Youkparigha, F.O. (2019). Larvicidal activity of fresh aqueous and ethanolic extracts of *Cymbopogon citratus* (DC) Stapf on Malaria Vector, *Anopheles gambiae*. *BAOJ Biotechnology*, 5:1-40.

Lentner, C., Lentner, C. and Wink, A. (1982). Students t- distribution tables. In Geigy scientific Tables Vol. 2. International Medical and Pharmaceutical information, Ciba-Geigy Limited, Basal, Switzerland.

Moola, A.K., Ayyadurai, T., Balasubramani, S., Vignesh, R., Mohan, P. K., Sathish, S. and Diana, R.K.B. (2023). Chemical composition and larvicidal activity against *Aedes aegypti* larvae from *Hyptis suaveolens* (L.) Poit essential oil. *Journal of Natural Pesticide Research, 3*:100018. https://doi.org/10.1016/j.napere.2022.100018

Pavela, R. (2009). Larvicidal property of essential oils against *Culex quinquefasciatus* Say (Diptera: Culicidae).*Industrial Crops and Products,* 30:311-315. https://doi.org/ 10.1016/j.indcrop.2009.06.005

Ravaomanarivo, L.H.R., Razafindraleva, H.A., Raharimalala, F.N., Asoahantaveloniaina, B., Ravelonandro, P.H. andMavingui, P. (2014). Efficacy of seed extracts of *Annona squamosa* and *Annona muricata* (Annonaceae) for the control of *Aedes albopictus* and *Culex quinquefasciatus* (Culicidae). *Asian Pacific Journal of*

Tropical Biomedicine, 4(10):798-806. https://doi.org/10.12980/APJTB.4. 2014 C1264

Roth, D., Henry, B., Mak, S., Fraser, M., Taylor, M., Li, M. and British Columbia West Nile Virus Surveillance Team, (2010). West Nile virus range expansion into British Columbia. *Emerging Infectious Diseases,* 16(8):1251-1258.https://doi.org/10. 3201%2Feid1608.100483

Shahat, M.A.M., El-Sheikh, T.M.Y., Hammad, K.M., Hasaballah, A.I. and Shehata, A.Z.I. (2020). Activity of *Otostegia fruticosa* (Lamiaceae) leaves extracts against lymphatic filariasis vector, *Culex pipiens* L. (Diptera: Culicidae). *Egyptian Academic Journal of Biological Sciences A. Entomology,*13(4):175-186. https:// doi.org/10.21608/eajbsa.2020.126835

Sharma, P., Mohan, L. and Srivastava, C.N. (2006 a). Phytoextract-induced developmental deformities in malaria vector. *Bioresource Technology,* 97:1599-1604.https:// doi.org/10.1016/j.biortech.2005.07.024

Sharma, P., Mohan,L. and Srivastava, C.N. (2006 b). Growth Inhibitory Nature of *Artemisia annua* Extract against *Culex autnauetesctetus* (Say). *Journal of Asia-Pacific Entomology,* 9(4):389395.

Shehata, A.Z., El-Sheikh, T.M., Shaapan, R.M., Abdel-Shafy, S. and Alanazi, A.D. (2020). Ovicidal and latent effects of *Pulicaria jaubertii* (Asteraceae) leaf extracts on *Aedes aegypti. Journal of the American Mosquito Control Association,* 36(3):161-166. https://doi.org/10.2987/20-6952.1

Shehata, A.Z.I. (2018). Feeding rate and reproductive performance of three mosquito species as influenced by different blood meal sources. *Egyptian Academic Journal of Biological Sciences A. Entomology,*11(6):77-84. https://doi.org/10.21608/eajbsa. 2018.26438

Shehata, A.Z.I. (2019). Biological activity of *Prunus domestica* (Rosaceae) and *Rhamnus cathartica* (Rhamnaceae) leaves extracts against the mosquito vector, *Culex pipiens* L. (Diptera: Culicidae). *Egyptian Academic Journal of Biological Sciences F. Toxicology & Pest control,* 11(1):65-73. https://dx.doi.org/10.21608/eajbsf. 2019.29343

Shehata, A.Z.I., El-Tabakh, M.A.M., Waheeb, H.O., Emam, D.E.M. and Mokhtar, M.M. (2022). Seasonal abundance and molecular identification of aquatic larvae of *Culex pipiens* L. and *Culex antennatus* Becker in Fayoum Governorate, Egypt. *Egyptian Journal of Aquatic Biology & Fisheries,* 26(6): 751-764. https://dx.doi.org/ 10. 21608/ejabf.2022.275615

Shehata, A.Z.I., Hammad, K.M. and Abdel-Samad, M.R.K. (2019). Toxicological and repellent effects of *Lantana camara* (Verbenaceae) and *Eucalyptus citriodora* (Myrtaceae) extracts against Rift Valley Fever Vector, *Culex antennatus* (Becker) (Diptera: Culicidae). *Journal of Nuclear Technology in Applied Science,*7(10):1-9. https://doi.org/10.21608/jntas.2019.54581

Shehata, A.Z.I., Labib, R.M. and Abdel-Samad, M.R.K. (2021). Insecticidal activity and phytochemical analysis of *Pyrus communis* L. extracts against malarial vector, *Anopheles pharoensis* Theobald, 1901 (Diptera: Culicidae). *Polish Journal of Entomology,* 90(4):209-222. https://doi.org/10.5604/01.3001.0015.6329

Toppozada, A., Abdallah, S. and El-Defrawi, M.E. (1966). Chemosterlization of larvae and adults of the Egyptian cotton leafworm, *Prodenialittura* by apholate, metepa and hempa. *Journal of Economic Entomology,* 59:1125-1128.https://doi.org/10. 1093/jee/59.5.1125

Vijayakumar, S. and Amirthanathn, A. (2014). Bioactivity of sea grass against the malarial fever mosquito *Culex quinquefasciatus*. *Asian Pacific Journal of Tropical Disease*, 4(4):287-291. https://doi.org/10.1016/S2222-1808(14)60574-0

World Health Organization, W.H.O. (2004). Integrated Vector Management: Strategic Framework for the Eastern Mediterranean Region 2004-2010. The WHO Regional Office for the Eastern Mediterranean, Cairo, 26 pp. https://apps.who.int/iris/handle/10665/119705