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Original Research Article

The Efficacy of Little Spurflower *Plectranthus parciflorus* Oil against Adult Mosquito (*Culex quinquefasciatus*)

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Abstract: The synthetic insecticides that are frequently used in mosquito control programs have developed insecticide-resistant mosquitoes. Thus, this study is aimed to evaluate the efficacy of essential oils from the arial part of leaf of *Plectranthus parciflorus* against adult *Culex quinquefasciatus*. The aerial parts of *P. parciflorus* was screened for its phytochemical constituents and used for adulticidal assay with stock solution of 5 g of the extract into 100 ml of water. A stock solution of the oil was prepared using 15% tween-40 solution as the diluent. Four different concentrations (62.5, 125, 250 and 500 ul/ml) of the extract with four replicates each. Forty (40) adults each of *Culex quinquefasciatus* were separately exposed to each extract concentration for 10, 20, 30, 40 and 50 mins. Rate of knockdown and mortality was recorded after each stipulated time intervals. The results revealed that the efficacy of the extract exposed to the samples increased with increased concentrations and duration of exposure. The knockdown and mortality rate of *P. parviflorus* oil against the *Cx. quinquefasciatus* samples were 200 (100%) and 148 (74%) respectively. This study revealed the susceptibility of adults *Cx. quinquefasciatus* to the extract as evident by the 50min LC_{50}, KD_{50} and KD_{95} values of 49.223 µL/mL, 6.726 µL/mL and 110.438 µL/mL respectively. Results obtained from this study suggest that the aerial parts of *P. parviflorus* is an effective constituent in the formation of pesticides or insecticidal of *Cx. quinquefasciatus*. Also, based on the exposure time, studies are yet to identify any eco-unfriendly attributes. Hence, it is a recommendable plant-based species with adulticidal potential against *Culex quinquefasciatus*.

Keywords: Plant-based insectides, Efficacy, *Plectranthus parciflorus* and *Cx. Quinquefasciatus*.

1.1. INTRODUCTION

Mosquitoes act as vectors for numerous disease-causing viruses and parasites, transmitting these pathogens from person to person without showing any symptoms themselves (Zapout USA, 2020). Diseases spread by mosquitoes include yellow fever, dengue fever, and chikungunya, which are primarily transmitted by *Aedes aegypti*. Dengue fever is the leading cause of fever in travelers returning from the Caribbean, Central and South America, and parts of South and Southeast Asia. The disease is spread through the bites of infected mosquitoes and cannot be transmitted directly from person to person. Severe cases of dengue can be life-threatening, but with proper medical care, the fatality rate is less than 1% (WHO, 2019).

Malaria, a parasitic disease caused by *Plasmodium* species, is transmitted by female mosquitoes of the *Anopheles* genus. Lymphatic filariasis (the main cause of elephantiasis) can be transmitted by various mosquito species (WHO, 2019). West Nile virus is a growing concern in the United States, although global statistics on cases remain unreliable (CDC, 2020). Dengue viruses pose a significant global health risk, with severe cases often requiring hospitalization. Symptoms of dengue include high fever, body aches, vomiting, and skin rashes. Severe dengue may lead to symptoms like vomiting blood, bleeding from the gums or nose, and abdominal pain (CDC, 2020). The common house mosquito, *Culex quinquefasciatus*, is a primary vector for these diseases (CDC, 2020).

Culex quinquefasciatus is a global mosquito species responsible for spreading several diseases. In Egypt, it is the main vector for bancroftian filariasis caused by *Wuchereria bancrofti* (Darwish and Hoogstraal, 1981), Rift Valley fever virus (Conley *et al*., 2014), and West Nile virus (Amraoui *et al*., 2012). Mosquito control in Egypt has become increasingly difficult due to the rise in resistance of *Cx. quinquefasciatus* to various insecticides. Zayed *et al*., (2016) noted signs of larval resistance to organophosphate insecticides and widespread adult resistance to organochlorine, pyrethroid, organophosphate, and carbamate insecticides. The World Health Organization (WHO) regularly calls for updated data on vector resistance and insecticide susceptibility to improve control strategies. However, there is an urgent need for environmentally friendly alternatives to chemical pesticides. Biological control methods, such as the use of mosquito fish, nematodes, and copepods, have shown promise in reducing mosquito populations (Sanad *et al*., 2013) and also microorganism (Merenini *et al.*, 2015a).

Lymphatic filariasis (LF) is caused by filarial worms and transmitted by mosquitoes (Cano *et al*., 2014). There are 23 mosquito species known to transmit filariasis, including the *Mansonia* genus (Tan *et al*., 2015). Filariasis transmission risk increases in habitats conducive to mosquito breeding, such as lowland areas, rice fields, swamps, and forests (Curtis and Graves, 2010). Due to its large lowland areas, Indonesia remains highly vulnerable to mosquito-borne filariasis (Kementerian, 2014).

Although there has been progress in mosquito control through the distribution of long-lasting insecticidal nets (LLINs) (WHO, 2011), more effort is required to increase coverage and reach remote communities. High coverage of insecticidal methods exerts strong selective pressure, promoting insecticide resistance in mosquito populations (Ranson *et al*., 2011). This highlights the importance of monitoring resistance, managing it effectively, and exploring diverse control strategies.

Targeting *Cx. quinquefasciatus* remains a key focus in disease control programs. Since insecticides are central to integrated vector management, developing safer and more effective alternatives is crucial (WHO, 2018). Using natural products to reduce mosquito populations at the larval stage offers potential benefits for vector control. Natural products with short environmental latency may reduce the risk of resistance development (Hardin and Jackson, 2009). Plants are increasingly being studied for their insecticidal properties since they pose less environmental harm. Since 1950, approximately 247 plant families have been identified to possess insecticidal toxins, including alkaloids, phenols, and terpenes (Silva Aguayo, 2004). Various organic and inorganic plant extracts, as well as plant-based secondary metabolites, have shown effectiveness in controlling mosquito populations (Lundberg, 2002).

Plants as medicinal and food ingredients are very important in the development of human civilization. The plants are direct or indirect impact to human health, so it’s to be carried out as new alternative therapeutic agents. The plants more than 7,000 species used as traditional medicinal. The plants efficacy is related to their bioactive compounds such as phenols, flavonoids, and tannins (Erdem *et al.,* 2015). Oils extracted from plants are widely studied and used as fragrances in cosmetics, food additives, agricultural protection agents, household products and medicines (Cheon *et al.,* 2009).

The genus Plectranthus (Lamiaceae) is represented by 14 species in India (Singh *et al*., 2001). It is an herb, locally called Lal aghada, used as a respiratory stimulant, vasoconstrictor, fever reducer, cardiac depressant and is also used for rheumatism, hemorrhage, mental retardation, snakebites as well as a general tonic (Lukhoba *et al.,* 2006). It is also used as an insect repellent (Lukhoba *et al*., 2006). Pharmacologically, it is reported to exhibit relaxant activity on smooth and skeletal muscles as well as cytotoxic and anti-tumor promoting activities (Lukhoba *et al*., 2006). Antimicrobial and bronchodilatory activities of the essential oil are reported (Sharma and Ali 1966). Previous phytochemical investigations Plectranthus have resulted in the isolation of fatty acids from seeds (Mahmood *et al.*, 1989) and b-sitosterol from hexane extracts of the whole plant (Desai *et al.*, 1977).

Various studies of plant oil extracts to control mosquitoes have been carried out, but limited on the “Control of *Culex quinquefasciatus* mosquito using *Plectranthus parciflorus* leaf”. Thus, the need to evaluate the efficacy of essential oils from the arial part of *Plectranthus parciflorus* against adult *Culex quinquefasciatus*.

2.0. MATERIALS AND METHODS

2.1. Collection and Identification of the Experimental Plants

The aerial parts of *Plecthranthus parciflorus* used in this study were provided and identified in the department of Plant Science and Biotechnology, Faculty of Science, Federal University of Lafia, Nasarawa State, Nigeria, and were kept in the herbarium specimens were prepared and kept for future reference.

2.2. Oils Extraction

The essential oils used for this study were obtained by hydrodistillation, using a Clevenger tight apparatus in the laboratory of the Department of Clinical Science, Faculty of Pharmacy, Federal University of Lafia, Nasarawa State, Nigeria. The aerial parts of *P. parciflorus* were washed and shade dried on the laboratory benches and later chopped into pieces and pulverized using a manual blender. The pulverized material was weighed (1500g) and introduced into a round bottom flask of a Clevenger apparatus. Four litres of water were added to the pulverized material in the Clevenger apparatus. The set up was allowed to boil for four (4) hours. At the end of 4 hours, the oil was collected through a tap into a glass sample bottle. The oil obtained was dried using sodium sulphate and stored at -4ºC for further use.

2.3. Preparation of Stock Solution and Test Oil concentration

For each oil, the stock solution was prepared by dissolving 1600µl of each oil in 2mls of 15% tween - 40 solutions to obtain 400µl/ml of each essential oil. From the stock solution of 400µl/ml of each oil, 1.5ml was dissolved in 4ml of tween-40 solution to obtain 500, 250, 125 and 62.5µl/ml of *P. parciflorus* by serial dilution.

2.4. Experimental Mosquito Species

Mosquito species used for this research was *Culex quinquefasciatus*. These mosquito specimens were harvested from their habitats within Lafia environs. Adult mosquitoes of each species were separately transferred into insect cages where they were fed and maintained on a 10% sucrose solution as described by the World Health Organization (WHO, 2005) and Onyido *et al.* (2009). They were left for 24 hours before the commencement of the experiment.

2.5. Knockdown and Insecticidal Testing

The knockdown and insecticidal potential of *P. parciflorus s*oils were separately tested against *Culex quinquefasciatus* in the Laboratory of the Department of Clinical Science, Faculty of Pharmacy, Federal University of Lafia, Nasarawa State, Nigeria. Federal University of Lafia, Nasarawa State. The experiment was conducted at an ambient temperature of 25 ± 2°C and 40 ± 5% Relative Humidity (RH). A stock solution of the oil was prepared using 15% tween-40 solution as the diluent. From the stock solution four different concentrations (62.5, 125, 250 and 500µl/ml) of *P. parciflorus* oils were used for the study in four replicates. The control which consisted of 1ml of 15% 10 also was replicated four times. Ten adult mosquitoes were exposed to each test concentrations and their respective replicates. The same was done for the control experiments. The adult mosquitoes were collected from the insect cage and transferred into different transparent WHO insecticide susceptibility test tubes using an aspirator.

The knockdown and insecticidal potential of the plant essential oils were then tested by separately impregnating Whatman No 1 filter paper with different concentrations of the oil, using a micropipette. The filter paper was then placed in the WHO approved Insecticide susceptibility test tubes as described by WHO, (2016). Observation of the knockdown and insecticidal effects of the oils on the adult mosquitoes were observed for 50 minutes at intervals of 10 minutes for both knockdown and mortality. The results obtained were recorded accordingly.

2.6. Statistical Analysis

The data were statistically evaluated by analysis of variance, followed by Duncan's multiple range test to examine the significant differences between treatments. The 5% level of probability was used in all statistical tests.

3.0. RESULTS AND DISCUSSION

3.1. Results

3.1.1. Percentage mortality of *Culex quinquefasciatus* using *Plectranthus parviflorus* oils

The percentage mortality rate of *Culex quinquefasciatus* with *Plectranthus parviflorus* oil was revealed in Table 1. The results revealed that the mortality rate increased with time and concentration. 62.5 and 125 (µL/mL) concentration levels of the oil had no potency on *Culex quinquefasciatus* in 10 mins application. Control treatment all showed 100% negative results irrespective of time. The response of the *Plectranthus parviflorus* oil tested against 40 samples of *Cx. quinquefasciatus* was respect to time (mins) and concentration levels of exposure. The rate of knockdown increased in mosquito species as the concentration of the oil increased with time which is presented in Tables 1. The oil was almost 100% potent against *Cx. quinquefasciatus*. 20 - 50 mins application of 62.5 – 250 (µL/mL) resulted to 100% knockdown rate in all the 200 samples of mosquitoes. The knockdown and mortality rate of *Plectranthus parviflorus* oil against 40 samples of *Cx. quinquefasciatus* was 200 (100%) and 148 (74%) respectively.

The $LC_{50,}$ KD_{50} and KD_{55} were values of the *Plectranthus parviflorus* oils was 49.223 µL/mL, 6.726 µL/mL and 110.438 µL/mL respectively.

Table 1: Percentage mortality of *Culex quinquefasciatus* vs *Plectranthus parviflorus*

Conc. (µL/mL)	No. of insects	Percentage mortality in minutes				
		10 (%)	20 (%)	30 (%)	40 (%)	50 (%)
62.5	40	0	6.5	30	60	80
125	40	0	15	40	65	90
250	40	7	35	72.5	80	100
500	40	30	65.5	95	90	100
Control	40	0	0	0	0	0

Table 2: Percentage Knockdown of *Culex quinquefasciatus* vs *Plectranthus parviflorus* in minutes

Conc. (µL/mL)	No. of insects	Percentage knockdown in minutes				
		10 (%)	20 (%)	30 (%)	40 (%)	50 (%)
31.25	40	35	70.5	100	100	100
62.5	40	65	90	100	100	100
125	40	30	100	100	100	100
250	40	85	100	100	100	100
Control	40	0	0	0	0	0

Table 3: 50minutes LC_{50} values of the *Plectranthus parviflorus* oils against *Culex quinquefasciatus*

Oils	LC_{50} values	KD_{50} values	KD_{95} values
Plectranthus parviflorus	49.223 µL/mL	6.726 µL/mL	110.438 µL/mL

3.2. Discussion

Cladophoraceae, *Asteraceae*, *Meliaceae*, *Rutaceae*, *Solanaceae* and *Lamiaceae*, are members of the plant families, that have been studied to possess larvicidal and adulticidal, repellent against different mosquito species (Sukumar *et al*., 2018). These results revealed that *Plectranthus parviflorus* was more effective than *Eryngium foetidum* on *Culex quinquefasciatus*. Among the 200 samples used for *Plectranthus parviflorus*, the percentage mortality of *Culex quinquefasciatus* was 74%. This means that about 148 samples died as a result of the exposure of the oil across different exposure periods. This study incorporates earlier findings of Govindarajan (2010), who reported that extract of *Tagetes patula* was less active and only 50 samples were killed at higher concentration (100 ppm).

The oil of *Plectranthus parviflorus* leaf extract tested for adulticidal activity *against Culex quinquefasciatus* with LC_{50}, KD_{50} and KD_{95} values of 49.223 µL/mL, 6.726 µL/mL, 110.438 µL/mL respectively. The results indicated that at concentrations of 62.5 µL/mL and 125 µL/mL, the oil showed no immediate effect (within 10 minutes), but mortality increased significantly with prolonged exposure. Complete knockdown was achieved within 20–50 minutes at concentrations of 62.5–250 µL/mL, suggesting a strong and rapid action of the oil against *Cx. quinquefasciatus*. The LC_{50} value of 49.223 µL/mL, KD_{50} value of 6.726 µL/mL, and KD_{95} value of 110.438 µL/mL reflect the high potency of *Plectranthus parviflorus* oil, comparable to other plant-based insecticides studied in recent research (Dias *et al*., 2023; Singh *et al*., 2021).

The knockdown percentage of the 200 samples was 74% for *Plectranthus parviflorus* which is over 160 samples knocked down. The results are due to the fact that a cold extraction method was used. Maceration has been reported to result in low yield but has the advantage of preserving the thermolabile components of the plant (Govindarajan *et al.*, 2010).

The aerial parts of *Plectranthus parviflorus* have reported some of the phytochemicals to be detected to have good adulticidal efficacy. The adulticidal potency of alkaloids, diterpenes, hydroxycinnamic acids, triterpenes, flavonoids and ferulic, sinapic acid, parientin have been reported to be present in *Plectranthus parviflorus* oil (Liu *et al*., 2012; Adefolalu *et al*., 2015; Perumalsamy *et al*., 2015). Thus, the adulticidal mortality rate of *Plectranthus parviflorus* reported in this study is attributable to the presence of these bioactive compounds. These compounds may have acted jointly or independently to contribute to their mortality and knockdown.

Reports on mosquito adulticidal activity of essential oils showed weak to moderate activities. For example, oil of *Lavandula officinalis* was reported to exhibit mortality to *Anopheles stephensi* (LC_{50} at 83.6 mg/L) and *L. angustifolia* to *Aedes albopictus* (LC_{50} at 250 mg/L) (Kumar and Dutta 1987). Composition of the *Eryngium foetidum* oil was found to contain Oleic acid, Stearic acid and Cetene as the major constituents (Sumitha *et al*., 2014).

The compound -2-dodecen-1-al is one of the major constituents of the oil of *Eryngium foetidum.* 2-dodecen-1-al (48.42 %) was found to be the most abundant compound in the essential oil of *E. foetidum*. Previous studies had confirmed the presence of 2- dodecenal in *E. foetidum* (Yarnell, 2004), thus it can be inferred that this compound leads to adulticidal activity on *Culex quinquefasciatus*. This demonstrates that it has natural insecticidal potentiality. Though reported to have a low potency on *Culex quinquefasciatus* larvae with an LC_{50} value of 956.013 µL/mL, which agrees with Koliopoulos *et al.*, (2010) and Merenini *et al.*, (2025b).

Syed *et al.*, (2012) reported that the potency of essential oils against mosquitoes may vary depending on plant species, plant parts, age of plant, solvent used in extraction and mosquito species tested. This may have also accounted for the variation in the susceptibility of the mosquito species to the extract. The mosquito species gave different results which dependent on the oil used. Mortality rate was noted to increase with increased concentration against period of exposure (mins). Furthermore, the rapid knockdown and high mortality rates seen in this study suggest that *Plectranthus parviflorus* oil could be integrated into mosquito control programs, particularly in regions where resistance to synthetic insecticides is widespread. The short environmental latency of plant-based insecticides may also help minimize the risk of resistance development, as reported by Silva *et al*., (2022).

3.3. CONCLUSION

Researchers are exploring plant-based adulticides as eco-friendly alternatives to chemical insecticides for mosquito control. *Plectranthus parviflorus* oil has shown significant adulticidal activity against *Culex quinquefasciatus*, highlighting its potential as a natural mosquito control agent. The oil is cost-effective, easy to handle, and safer for the environment and human health. Combining plant-based oils could provide a multi-component alternative to synthetic pesticides. Further research is needed to isolate and characterize the active compounds in *P. parviflorus* oil for industrial use.

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