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Mosquito adulticidal activities of crude extracts from above ground part of *Laggera pterodonta* (D.C) Sch.Bip and *Laggera aurita* (D.C) Sch.Bip against malaria vector, *Anopheles gambiae*

Dantanko, Fatima[1]*, Ojiego, O. Beatrice[1], Ibrahim, Halima[2], Bello Zainab[1], Abiamere Cynthia[1], Malann, Y. D.[3] and Dantanko, Hadiza[4]

[1]Environmental Biotechnology and Bioconservation Department, National Biotechnology Development Agency, Abuja, Nigeria.
[2]Department of Biological Science, Kaduna State University, Nigeria.
[3]Biological Sciences Department, University of Abuja, Nigeria.
[4]Agricultural Biotechnology Department, National Biotechnology Development Agency, Abuja, Nigeria.

*Corresponding author. Email: fatimadantanko@yahoo.com; Tel: +2348037013662.

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ABSTRACT: Mosquitoes are small, midget flies that transmit harmful infections such as malaria, yellow fever, and dengue fever in Nigeria. Its control using a synthetic insecticide is becoming increasingly difficult due to high cost, persistence in the environment, and development of resistance by the mosquitoes. The aim of this study is to evaluate the adulticidal activity of crude ethanolic extract of the above ground parts of *Laggera pterodonta* and *Laggera aurita* against adult *Anopheles gambiae*. Dried *Laggera pterodonta* and *Laggera aurita* were individually macerated with 70% ethanol to obtain the crude extract. Phytochemical screening revealed the presence of saponin, flavonoids and terpenoids in both *Laggera pterodonta* and *Laggera aurita* while tanins and alkaloids were present in *Laggera aurita* only. The extracts were tested against 4-5 days old laboratory-reared adult *Anopheles gambiae.* The findings revealed that the adulticidal lethal concentration LC_{50} of *L. pterodonta* crude extract was 4553 mg/L while the LC_{50} of *L. aurita* was 5582 mg/L. The result suggests the extracts are potential natural insecticides against adult *An. gambiae* mosquitoes.

Keywords: Adulticide, crude extract, *Laggera pterodonta*, *Laggera aurita*, mosquito.

INTRODUCTION

Mosquitoes are small, midget flies that transmit harmful infections such as malaria, yellow fever, and dengue fever in Nigeria. Malaria infection remains a major public health challenge and socio-economic development hindrance in Nigeria. It is a risk for 76% of Nigeria's population (USAID, 2021). It is a serious disease affecting children and adults but its consequences are graver among children below five years and pregnant women (Jimoh *et al*., 2007). Malaria is caused by the *Plasmodiun* parasite, which is spread by the bite of infected female mosquitoes to humans. *Anopheles gambiae mosquito* is the primary vector of malaria in Nigeria.

Vector control using an insecticide is one of the vital components of World Health Organization WHO's Global Malaria program (World Health Organization, 2021). However, extensive synthetic chemical insecticides-based intervention measures for the control of malaria vectors have resulted in adverse environmental effects, increased public concern about the safety of many chemical products, and the development of insecticide resistance

(Edriss *et al*., 2013; Singh *et al*., 2001). Insecticide resistance is increasingly becoming a problem for many vector control programs. Resistance of vector mosquitoes to various classes of insecticide has been reported in Nigeria (Muhammad *et al*., 2021; Chukwuekezie *et al*., 2020; Habibu *et al*., 2017; Djouaka *et al*., 2016). Therefore, systematic screening of plants may result in the discovery of novel effective compounds which would tackle the problem of insecticide resistance.

In the last few years, several researchers started the development of plant based insecticides due to their excellent larvicidal and adulticidal properties to replace synthetic insecticides which are associated with wide spread resistance and non-biodegradable in mosquito control programs. The genus *Laggera* belongs to the family Asteraceae and consists of about 20 species. Plants in the genus *Laggera* are aromatic and mostly found growing as weed in Africa and Asia. *Laggera pterodonta* and *Laggera aurita* are the two common species found growing in Nigeria. They are mostly used interchangeable for the treatment of pediatric malaria, epilepsy, fever, pain, nasal congestion, and indigestion (Egharevba *et al*., 2010; Magajia and Sani, 2018). The mosquito repellent and oviposition deterrent activities of the acetone crude extract of the whole plant of *L. aurita* have been reported (Singh and Mittal, 2015). The activity of the crude extracts, fractions, and essential oils of *L. pterodonta* and *L. aurita* against the larvae of *Anopheles gambiae* has been reported (Dantanko and Malann, 2019; Dantanko *et al*., 2021). Also, Dantanko *et al*. (2022) reported the adulticidal activity of the essential oil of both *L. pterodonta* and *L. aurita* against *An. gambiae*. The present study aims to evaluate the adulticidal activity of the crude extract of the above ground parts *L. pterodonta* (D.C) Sch.Bip and *L. aurita* (D.C) Sch.Bip against female *An. gambiae.*

MATERIALS AND METHODS

Plant collection

Above ground parts of the plants (*Laggera aurita and Laggera pterodonta*) were collected from Chaza village in Suleja, Niger State Nigeria. They were identified and authenticated by a botanist in the Herbarium Unit of Medicinal Plant Research and Traditional Medicine Department, National Institute for Pharmaceutical Research and Development (NIPRD). *L. aurita* was assigned voucher specimen number of NIPRD/H/6977and NIPRD/H/6978 was assigned to *L. pterodonta*. The plants were shade dried at room temperature (27±2°C) for two (2) weeks. The particle size was reduced by pounding using mortar and pestle (Sofowora, 1982).

Preparation of crude ethanol extracts

A weight of 1000 g (1 kg) of each dried plant was macerated with 6 litres of 70% ethanol for 24 hours at room temperature. The extract was filtered with muslin cloth and then using vacuum filtrator (Charles Austen pumps innova 20). The solvent was removed using rotary vacuum evaporator (RE 100) at 30°C and a thick solution of the filtrate was obtained. It was then transferred to a stainless-steel bowl and concentrated in a water bath at 40°C. This afforded 49.46 g of crude extract of *L. pterodonta* and 50.98 g of *L. aurita*. After complete evaporation of the solvent, the concentrated extract was collected in a vial and stored at room temperature (Ekpendu *et al*., 2000).

Phytochemical analysis

Samples of the test plant materials were analyzed for alkaloids, flavonoids, tannins, saponins and terpenoids using the standard procedures as described by Harborne (1998) and Sofowora (1982) at NIPRD, Abuja.

Collection and rearing of *Anopheles gambiae*

Mosquito larvae were collected from plastic containers containing water that was placed outside. It was taken to the Biology Laboratory of the Department of Biological Sciences, University of Abuja and identified - using keys provided by Gillies and De Meillon (1968). The identified larvae were transferred into a beaker containing water the beaker was then covered with net (polynestrene). Larvae were maintained by feeding them with larval food (ground fish). Within 2-3 days, the larvae were transformed into pupae and then adults emerged and were found hanging on the net.

Adulticidal test

Appropriate concentrations of different plant extracts were dissolved in 2.5 mL Dimethyl sulphoxide (DMSO) at the Biology Laboratory, University of Abuja. The solution was impregnated on Whatman no.1 filter papers (size 12 x 15 cm^2) with the help of the pipette in a plastic container and an untreated net covering the beaker was also impregnated (Gimba, 2016). The plant extracts were evaluated at five concentration; 1000, 2000, 3000, 4000 and 5000 mg/mL. Equal amount of solvent (DMSO) was added on the filter paper without the extract and kept as control. The papers were allowed to dry overnight at room temperature under shade before testing. The dried filter papers were placed in 250 ml glass beakers similar to the Centers for Disease Control and Prevention (CDC) glass bottle bioassay of insecticides. A total of 20 laboratory reared adult *Anopheles gambiae* mosquitoes were gently transferred into each beaker. The experiment was carried out in triplicate. Therefore, a total of 80 mosquitoes were assayed for each plant extract and one batch as a negative control group for each concentration. Adult food, 10%

sugar solution was provided to both the experimental and the control adults. Mortality was recorded at 24 hours intervals. Mosquito mortality was recorded as dead if it was lying on its back or side and was unable to maintain flight after a gentle tap on the body (Dantanko *et al*., 2022).

Statistical analysis

SPSS 20.0 version package was used for analyzing the data. Data from mortality and the effect of concentrations were subjected to analysis of variance (ANOVA). Probit analysis was used to determine the LC_{50} and LC_{90} at 95% confidence limits of upper confidence limit and lower confidence limit.

RESULTS AND DISCUSSION

Phytochemical constituents of the crude extract of *Laggera pterodonta* and *Laggera aurita*

The qualitative determinations of some phytochemicals present in the crude extracts of *Laggera pterodonta* and *Laggera aurita* are presented in Table 1. The result shows that saponins, flavonoids and terpenoids were present in both *L. pterodonta* and *L. aurita* while alkaloids and tannins were present in *L. aurita* only. Phytochemical screening provides basic information about the different classes of secondary metabolites present in a plant (Chintem and Nzelibe, 2015).

The result of phytochemicals of *L. pterodonta* in this study is in agreement with that of Ikyenge *et al*. (2019) who also reported saponin, flavonoid and terpenoids present in *L. pterodonta*. The presence of these phytochemicals could be attributed to the susceptibility of the plant extracts against mosquito adult. Saponins are secondary plant metabolite known to be toxic to harmful insects, retardation in development and also known for its anti-feeding properties (Gutierrez *et al*., 2014; Udebuani *et al.*, 2015). Flavonoids have been reported as effective insecticide against mosquitoes by altering their normal body functions (Kotkar *et al*., 2002). Alkaloids and tannins are also known to possess pesticidal properties (Azmathullah, 2011). Therefore, it is not possible to attribute the adulticidal activity to one principle phytochemical among those detected in the phytochemical screening.

Adulticidal activity of crude extracts of *L. pterodonta* and *L. aurita*

In Figure 1, the crude extract of *L. pterodonta* recorded the highest mortality of 55.83% at a concentration of 5000 mg/L while the lowest mortality of 0.83% was recorded at a concentration of 1000 mg/L. The crude extract of *L. aurita* showed weak adulticidal activity, with a mortality of 44.18% at the highest concentration of 5000 mg/L. No mortality was recorded at the concentration of 1000 mg/L. Different phytochemicals have been reported to have weak to strong adulticidal activity against mosquito species. Although, all phytochemicals reviewed were present in *L. aurita,* the potency of the active constituents against adult *Anopheles gambiae* may have been masked by other less active or completely inactive constituents in *L. aurita*. Hence, the higher activity of the extract of *L. pterodonta*. Dantanko *et al.* (2021) reported saponin fraction of *L. aurita* as weak mosquito larvicde in contrast with Chaieb (2010) who reported strong larvicidal activity of crude saponin of *Cestrun paraqui*.

Table 1. Phytochemical constituents of *Laggera pterodonta* and *Laggera aurita crude extracts.*

Phytochemicals	*L. pterodonta*	*L. aurita*
Saponins	+	+
Flavonoids	+	+
Tannins	-	+
Akaloids	-	+
Terpenoids	+	+

+ = present, - = absent.

The adulticidal results of the crude extracts recorded in this study are in agreement with Elango *et al.* (2012) who reported extracts of *Eclipta prostate* (Asteraceae), *Tagete erecta* (Compositae), *Aegele marmelos* (Rutaceae), *Andrographis paniculata* (Acanthaceae) and *A. lineata* (Acanthaceae) as being toxic to adult *Culex tritaeniorhynchus* mosquitoes. Research conducted on the crude extracts of *Ocimum canimum*, *O. gratissimum*, *Chromolaena odorata* and *Datura stramonium* also found them toxic to the adult *An. gambiae* (Afolabi *et al*., 2018).

Lethal concentration of extracts against adult of *Anopheles gambiae*

Table 2 shows the lethal concentration of the crude extracts of *L. pterodonta* and *L. aurita*. The crude ethanol extracts of both *L. pterodonta* (LC_{50} 4553 mg/L) and *L. aurita* (LC_{50} 5582 mg/L) were not highly toxic to the adult *An. gambiae* in this study when compared to their toxicity against the larvae of *An. gambiae* (*L. pterodonta* LC_{50} 2805 mg/L and *L. aurita* LC_{50} 4281 mg/L) as reported by Dantanko *et al.* (2021) and Dantanko and Malann (2019). In testing for adulticidal activity, extracts of *Gloriosa superba, Ricinus communis, Tridax procumbens, Solanum tribulatum, Anisomeles malabarica* and *Ocimum basilicum* also exhibited adulticidal potency on *An. stephensi* with an LD_{50} value of 120.17, 108.77, 127.22, 163.11, 118.27 and 93.02 µg/L respectively (Zahir *et al*., 2010).

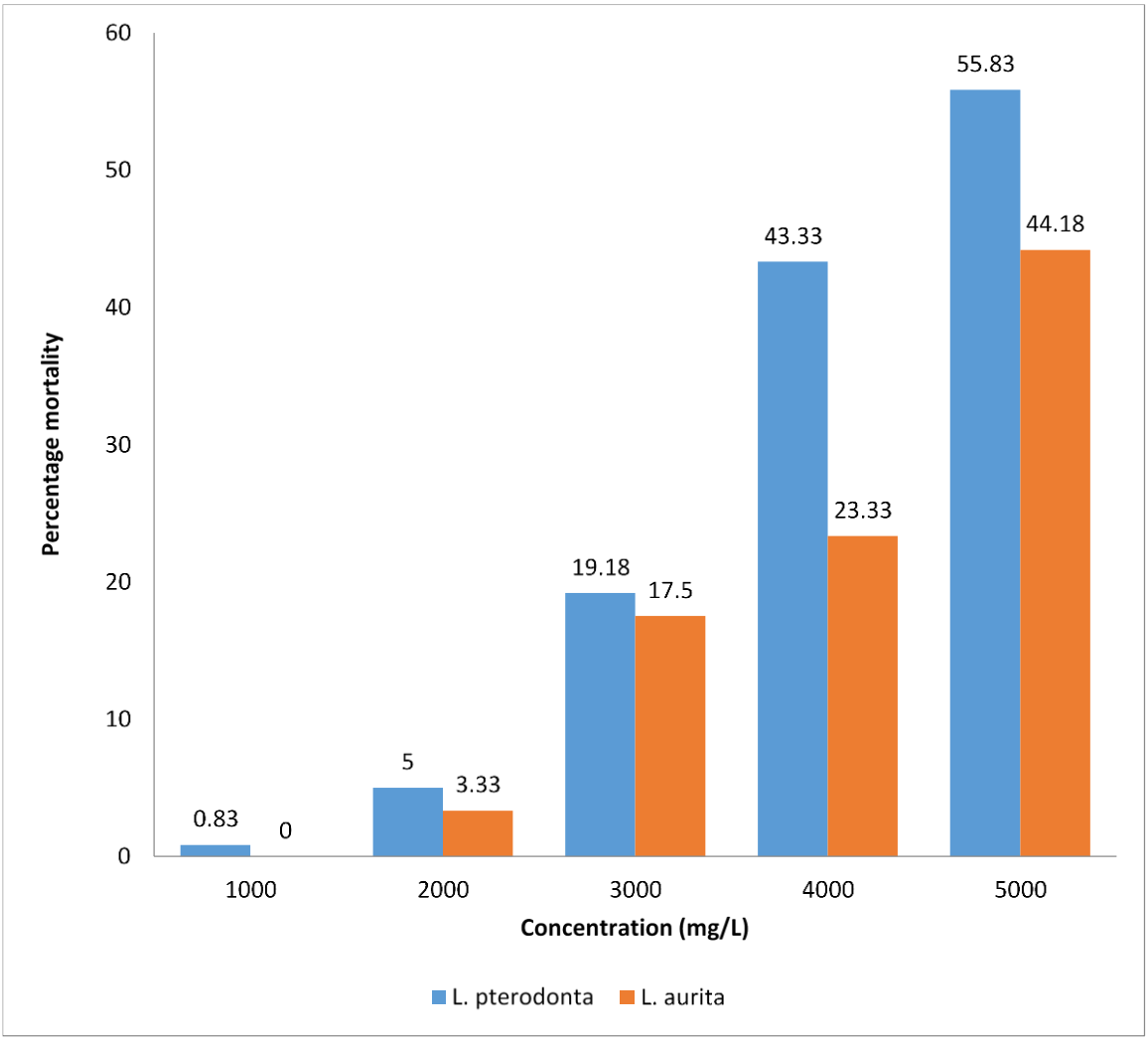

Figure 1. Adulticidal activity of crude extracts of *L. pterodonta* and *L. aurita.*

Table 2. Lethal concentration LC_{50} of crude extracts against adult *Anopheles gambiae* at 95% confidence interval.

Crude extracts	LC_{50} (LB- UB) mg/L	LC_{90} (LB- UB) mg/L
L. pterodonta	4553 (4198-5077)	8493 (7052-11709)
L. aurita	5582 (4980-6598)	11663 (8919-21263)

Conclusion

The obtained result shows that *L. pterodonta* and *L. aurita* has potential to be developed as an insecticide against *Anopheles gambiae*. The mosquitocidal activities were attributed to potent phytochemicals present in the extracts. However, studies aiming at isolating and identifying active compound(s) are in progress. Also, evaluation of the extracts on other species of mosquitoes is being considered. Such findings would offer an opportunity for developing newer more selective, biodegradable and natural mosquitocidal compounds as alternatives to rather expensive and environmentally hazardous inorganic insecticides.

Recommendation

Based on the findings of this work, the following recommendations are put forward:

1. Further studies should be carried out on the effect of *Laggera pterodonta* and *Laggera aurita* on other species of mosquitoes because mosquito responses to larvicides and adulticides vary within and among species.
2. Studies should be carried out to identify and isolate the mosquitocidal compound(s) present in the extracts for product development.

CONFLICT OF INTEREST

The authors declared that there is no conflict of interest.

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