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Assessment of the Larvicidal Efficacy of the Hexane-Leaf-Extracts of Selected Tropical Plant Species

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Abstract: The proliferation of mosquito vector in tropical countries has increased the prevalence of malaria with high morbidity and mortality burden. This research is concerned with the biolarvicidal efficacies of Cassia alata, Microdesmis puberula, and Spilanthes filicaulis extracted with hexane solvent, and screened against mosquito larva. Results on the negative control had no mortality on the larvae compared to the positive control, which had total mortality (p<0.05). The C. alata plant extracts was the most active with LC50 value of 25.26 ppm, followed by; M. puberula (32.83 ppm), and S. filicaulis (36.46 ppm). Based on the outcome of the larvicidal bioassay, Hexane-leaf-extracts of all plants can be recommended for the formulation of biolarvicide due to their efficacies.

Keywords: Mosquito; Malaria; Biolarvcide; Methanolic-extract

I. Introduction

Malaria diseases are a major problem ravaging the tropical county. Statistically, malaria was reported to affect over 3.5 billion persons annually (Ohimain *et al.*, 2014). It was also reported in literature that there are over 100 countries, with about 700 million incidence of malaria, especially tropical Region (Okumu *et al.*, 2007). Malaria disease has been reported in the many developing countries in especially in Africa.

There are diverse species of mosquitoes that exist in nature, but around 30 - 40 species were reported to be carriers of the plasmodium parasite (Ghosh *et al.*, 2007). In Africa, the most endemic transmitter of malaria is the female species of *Anopheles gambiae* and *An. Arabiensis* (Okumu, *et al.*, 2007; Angaye, *et al.*, 2014a; 2014b; Hamza *et al.*, 2014; Owoeye *et al.*, 2016).

According to Angaye (2015), the therapeutic efficacy of plant is not farfetched because plant produce elaborate metabolites as their genetic makeup, defence mechanisms and otherwise. As reported by Devappa, *et al.*, (2010), tannins produced by some plants have ability to retard the palatability, nutrient absorption and growth rate of predators. These metabolites found in plant, these includes about 10,000 and 25,000 alkaloids and terpenes respectively (Cheeke, 1998).

There are several compounding factors responsible for plant efficacies as antioxidant; this include age of the plant, location, season and even ability of the plant to withstand harsh environment (Devappa *et al.*, 2010; Angaye, 2015), or even the part of the plant (root, stem, fruits, leaves, and seeds) and/or applied solvent used for the bioassay (Angaye, 2015). Therefore, this investigation on the larvicidal efficacies of the hexane extracts of *C. alata*, *M. puberula*, and *S. filicaulis*.

II. Material and Method

2.1 Collection and Preparation of Plant Samples

The fresh leaves of *C. alata*, *M. puberula* and *S. filicaulis* were collected from vegetation around Wilberforce Island in Southern Ijaw Local Government Area of Bayelsa State, Nigeria. The plants were identified, washed and shade-dried for 7 days. The shade-dried plants were

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placed in oven at 50°C for 30 minutes (Angaye et al., 2017a; Angaye et al., 2017b), and powdered with electric blender.

2.2 Extraction Process

Three hundred grams (300 g) of the powdered leaves of each plants were weighed using Satoric AG Gottingen Electronic weighing balance. The weighed powdered leaves were respectively macerated in 500 ml of the Hexane solvent for 72 hours and filtered into conical flask using whatman No.1 filter paper (Azoro, 2000). The filtrates distinctly extracted using a rotary evaporator at 60°C.

2.3. Mosquito Larva Collection

Mosquito Larvae used for this bioassay were cultured in the wild using methods as described by some authors (Rai et al., 2004; Okumu *et al.*, 2007; Ohimain *et al.*, 2014), with slight modification. Plastic containers and automobile tyres half-filled with stagnant water, and sand were kept in vegetation of conspicuous breeding sites. Prior to the laboratory bioassay, the larvae were placed on enamel tray and acclimatized to laboratory condition.

2.4. Experimental Setup

A minimum of 10 larvae, were distinctly placed in a 500 ml solution of the methanolic-extract at varying concentrations, in a 24-hour static non-renewal test. The bioassay was performed with the standard of the World Health Organization guidelines (Dibua *et al.*, 2013). Mortality rates (%), of larvae were recorded after the period of exposure (24 hours). A concentration of 1 ppm of Dipex pesticide was used as the positive control, while 500 ml of distilled water was used as the negative control. The larvicidal screening protocols were two-phased, involving the rapid and final Screening (Agboola *et al.*, 2011).

2.5 Statistical Analysis

The data for mortality rates were expressed as mean± standard deviation using version 20 of SPSS statistical package. A one-way analysis of variance was used to carry out the statistical analysis, while Duncan multiple range test was used to determine the source of observed difference using SPSS Version 20.

III. Result and Discussion

Results of mortality rates for leaf Hexane extracts of the three plants is presented in table 2. For the *Cassia alata* extract, the mortality rates ranges from 26.66 - 100.00% significantly (p<0.05), with minimal and total mortalities at concentrations of 10 and 70 ppm respectively. In addition, the positive control had total mortality at concentration of 10 ppm, compared to the negative control that showed no mortality rate (Table 1). Furthermore, results of the larvicidal efficacy of the *C. alata* hexane extract were demonstrated with LC₅₀ value of 25.26 ppm (Figure 1).

Table 1. Biolarvicidal Mortality rates of Hexane-leaf-extracts

Tuble 1. Biolar violati infortanty faces of florance lear extracts				
Mortality Rates (%)				
C. alata	M. puberula	S. filicaulis	Positive	Negative
$0.00\pm0.00a$	$0.00\pm0.00a$	$0.00\pm0.00a$	$0.00\pm0.00a$	$0.00\pm0.00a$
26.66±11.54b	$13.33 \pm 5.77b$	$10.00\pm0.00b$	100.00±0.00e	$0.00\pm0.00a$
36.66±5.77bc	20.00±0.00c	16.66±5.77b	100.00±0.00e	$0.00\pm0.00a$
43.33±5.77cd	30.00±0.00d	26.66±11.54c	100.00±0.00e	$0.00\pm0.00a$
53.33±11.54d	$40.00\pm0.00e$	$30.00\pm0.00c$	100.00±0.00e	$0.00\pm0.00a$
$70.00\pm10.00e$	56.66±5.77f	46.66±5.77d	100.00±0.00e	$0.00\pm0.00a$
96.66±5.77f	$90.00\pm10.00g$	73.33±11.54e	100.00±0.00e	$0.00\pm0.00a$
100.00±0.00f	100.00±0.00g	$93.33 \pm 5.77 f$	100.00 ± 0.00 e	$0.00\pm0.00a$
	C. alata 0.00±0.00a 26.66±11.54b 36.66±5.77bc 43.33±5.77cd 53.33±11.54d 70.00±10.00e 96.66±5.77f	C. alata M. puberula 0.00±0.00a 0.00±0.00a 26.66±11.54b 13.33±5.77b 36.66±5.77bc 20.00±0.00c 43.33±5.77cd 30.00±0.00d 53.33±11.54d 40.00±0.00e 70.00±10.00e 56.66±5.77f 96.66±5.77f 90.00±10.00g	Mortality Rates (%) C. alata M. puberula S. filicaulis 0.00±0.00a 0.00±0.00a 0.00±0.00a 26.66±11.54b 13.33±5.77b 10.00±0.00b 36.66±5.77bc 20.00±0.00c 16.66±5.77b 43.33±5.77cd 30.00±0.00d 26.66±11.54c 53.33±11.54d 40.00±0.00e 30.00±0.00c 70.00±10.00e 56.66±5.77f 46.66±5.77d 96.66±5.77f 90.00±10.00g 73.33±11.54e	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$

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80 ppm 100.00±0.00g 100.00±0.00f 100.00±0.00f 100.00±0.00e $0.00\pm0.00a$ 90 ppm 100.00±0.00f 100.00±0.00g 100.00±0.00f 100.00±0.00e $0.00\pm0.00a$ 100 ppm 100.00±0.00f $100.00\pm0.00g$ $100.00\pm0.00f$ 100.00±0.00e $0.00\pm0.00a$

Data expressed as mean±standard deviation, differences in alphabet indicates significance in mortality

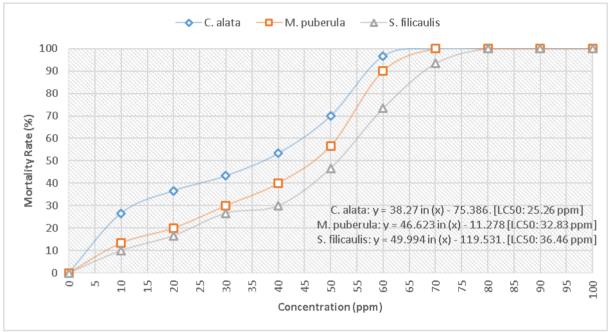


Figure 1. Biolarvicidal dose-response for Hexane-leaf-extracts of selected plant species

Results of the *M. puberula* bioassay showed that mortality rates ranges from 13.33 - 100.00%, with significant difference (p<0.05). While mortality rates increased as concentration increases (Table 1). The minimal mortality rate was reported at concentration of 10 ppm, while the total mortality was at 70 ppm. In addition, the activity of *M. puberula* hexane extract was demonstrated with LC₅₀ value of 32.83 ppm (Figure 8). The *S. filicaulis* biolarvicidal bioassay showed that mortality rate ranges from 10.00 - 100%, with significant increase in mortality rate and concentration increases (Table 4.2). The minimal mortality rate was reported at concentration of 10 ppm, while the total mortality rate was displayed at 80 ppm. In addition, the no adverse effect level was at 0.00 ppm (Table 4.2). The biolarvicidal activity of hexane extract of *S. filicaulis* was demonstrated with LC₅₀ value of 36.46 ppm (Figure 1).

Results reported for hexane extracts of the current study are comparable to results by other authors. Comparatively, the larvicidal activities of the hexane extracts of the bark (11.02 ppm) and root (28.08 ppm) *Azadirachta indica* were (Angaye *et al.*, 2014a), induced higher activities than hexane extracts of the 3 plants in the current. The larvicidal efficacies of the hexane leaves of Niger Delta mangrove plants against *Anopheles gambiae* was reported with activities for *Rhizophora mangle* (275.63 ppm), *R. racemosa* (225.00 ppm), *A. germinans* (250.50 ppm), and *L. racemosa* with value of 308.50 ppm (Angaye *et al.*, 2014b); these values were higher than values reported in the current study for all three plants.

The larvicidal activities of solvent extracts of *Hyptis suavolens* (76.25 ppm) and *Ocimun sanctum* (97.25 ppm) against mosquito was reported by Ohimain *et al.*, (2015), with higher LC₅₀ values than the current study. Sakthivadivel and Daniel (2008), reported higher LC₅₀ values (62.29 ppm) than values of the current study using petroleum ether solvent extract of *J. curcas* leaves against *An. stephensi*. The activities of these plants were due to presence of several phytochemicals (Ohimain *et al.*, 2015). Huge varieties of bioactive phytochemical have been previously reported in *C. alata* (El-mahmood and Doughari, 2008; Eliakim-Ikechukwe *et al.*, 2013; Okooboh and Gambo, 2013; Raji *et al.*, 2015; Ugbogu *et al.*, 2016;), *M. puberula* (-

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Akpanyung et al., 2013; Gbadamosi and Oloyede, 2014; Ndam et al., 2014; Eboh et al., 2017) and S. filicaulis (Wahab et al., 2013; Ilondu et al., 2014; Fonkeng et al., 2015; Eboh et al., 2017).

IV. Conclusion

This study investigated the biolarvicidal potential of the Hexane extracts of *C. alata*, *M. puberula*, and *S. filicaulis*. Fortunately, all plants demonstrated significant larvicidal efficacies with the *C. alata* extract demonstrating the highest efficacy, followed by *S. filicaulis* and *M. puberula*. Due to their efficacies, Hexane extracts of these plants, are hereby recommended for the formulation of larvicidal agents for the control of malaria.

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