Eurasia Specialized Veterinary Publication

Entomology Letters

ISSN:3062-3588

2021, Volume 1, Page No: 31-36

Available online at: www.esvpub.com/

Exploring the Toxic Effects of *Acacia nilotica* Fruit Water Extract on Mosquito Larvae: A Laboratory Investigation

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ABSTRACT

Mosquitoes are responsible for transmitting deadly pathogens, causing millions of fatalities annually. Efforts to improve mosquito control are a constant global challenge. Chemical control remains a dominant approach in vector management due to its rapid effectiveness, especially during outbreaks. However, the development of resistance to chemical insecticides has led to a resurgence in mosquito populations. Plants present a promising alternative for mosquito control solutions. This study investigates the toxic effects of aqueous extract from *Acacia nilotica* fruits on mosquito larvae under controlled laboratory conditions. This experimental study investigated the toxicity of aqueous extract of *Acacia nilotica* fruits on mosquito larvae. The fruit extract was dried, powdered, and strained to make a stock solution. A series of concentrations ranging from 0.0125-2% were applied to mosquito larvae, which were exposed to the extract for 24 hours. The lethal concentration at 50% (LC_{50}) was determined. After repeating the experiment 3 times, the mean result was calculated. Mortality rates ranged from 75% to 100%. The average LC_{50} in the three trials was 0.004%. Most of the mosquito larvae were identified as *Culex* species. The aqueous extract of *Acacia nilotica* shows significant toxicity against mosquito larvae and shows potential as an effective tool for mosquito control. Further research is needed to isolate the active compounds responsible for their toxicity and to explore additional solvent extractions and plant sources from different soils.

Keywords: Water extract, Mosquito larvae, *Acacia nilotica*, Toxic effects

Received: 20 May 2021
Revised: 30 August 2021
Accepted: 10 September 2021

How to Cite This Article: Saha HK, Das S. Exploring the Toxic Effects of *Acacia nilotica* Fruit Water Extract on Mosquito Larvae: A Laboratory Investigation. Entomol Lett. 2021;1:31-6.
https://doi.org/10.51847/VHGR6Tst1s

Introduction

Over two billion people, primarily in tropical regions, are at risk of diseases transmitted by mosquitoes, including malaria, dengue, hemorrhagic fever, and filariasis [1, 2]. These diseases predominantly affect the impoverished populations in tropical areas. Dengue infects an estimated fifty million people annually [3], while malaria significantly hampers Africa's economic development, further entrenching cycles of poverty [4]. The disease leads to around 300-500 million cases and over one million deaths each year [5]. The pathogens responsible for these illnesses are spread through the bites of blood-feeding mosquitoes in tropical and subtropical climates [6]. Mosquito genera such as *Culex*, *Aedes*, and *Anopheles* play a key role in transmitting diseases to humans. Despite the availability of effective control measures, mosquitoes remain a major public health concern, particularly in Africa. Many people in the region continue to be exposed to mosquito bites due to the widespread distribution of mosquito larvae in humid environments like flood zones and rice fields [7-10]. Modifying these breeding habitats could help reduce mosquito populations and disrupt disease transmission [11-13]. One effective strategy for reducing mosquito numbers before they reach adulthood is larviciding. While pesticides are highly efficient in this regard, their use comes with significant risks, including toxicity to humans and non-target animals. The overuse of chemical insecticides has led to resistance in mosquito populations, diminishing control effectiveness and causing environmental pollution, which in turn leads to bioaccumulation in the food chains and contamination. This study investigates the toxic effects of aqueous extract from *Acacia nilotica* fruits on mosquito larvae in controlled laboratory conditions.

Materials and Methods

Design

A laboratory-based experiment was conducted to examine the toxic effects of the aqueous extract from *Acacia nilotica* on mosquito larvae over a period from October 2017 to 2019. The research was carried out at the Faculty of Medical Laboratory Sciences, Al Neelain University. Ethical approval was obtained from the Institutional Review Board of the faculty before starting the study. To ensure accuracy, a trained entomologist verified both the species identification and the health status of the larvae before their use in the trials.

Collection of larvae

Mosquito larvae were gathered from Khartoum Bahry (Alfaky Hashem village) in Sudan and transferred to the laboratory with great care to prevent any drowning, the larvae were examined to identify their species and assess their viability before initiating any laboratory procedures.

Preparation

The fruits of *Acacia nilotica* were sourced from the market, dried in a shaded area, and finely powdered using an electric blender. A 2% (W/V) stock solution was prepared by mixing 20 grams of powdered fruit into 800 ml of distilled water and left to sit at room temperature (37 °C) for 24 hours. The mixture was then strained, and the volume adjusted to 1000 ml with additional distilled water, resulting in a stock solution of 20 grams per liter, equivalent to 20,000 ppm. Serial dilutions of this solution were prepared to create concentrations of 10,000, 5,000, 1,000, 500, 250, and 125 ppm.

Exposure of larvae to extract

For testing, 20 larvae were placed into small containers, each holding 50 ml of one of the prepared concentrations (20,000, 10,000, 5,000, 1,000, 500, 250, or 125 ppm). A control group was set up with 20 larvae in 50 ml of distilled water. After 24 hours, the larvae were observed, and mortality rates were recorded by counting both dead and alive larvae. Larvae were known dead if they settled at the bottom of the container without movement. All experiments were conducted at room temperature (30-37 °C) with the aid of a mercuric thermometer and was repeated 3 times.

Materials

The materials utilized in the study included 1000 ml plastic dishes, strainers, flasks, measuring cylinders, an electric blender, graph paper, and a calculator.

Analysis

Mortality data was converted into probit units and plotted against the logarithmic concentrations of the aqueous extract.

Results and Discussion

The experimental data was analyzed using analysis of variance (ANOVA) and log-probit transformation. Due to the unavailability of logarithmic graph paper, a graph was used for visualization. The data for each trial was recorded separately. The average mortality rate of mosquito larvae exposed to the *Acacia nilotica* aqueous extract across the 3 trials ranged from 75-90%, with probit values falling between 5.67 and 6.28. The concentrations tested ranged from 125 ppm to 5000 ppm. The LC_{50} values for the 3 trials were 0.0051, 0.004, and 0.004, with the overall average LC_{50} being 0.0044 **(Figures 1-4, Tables 1-4)**.

Table 1. Trial 1, Khartoum, 2017

PPM	Conc. (%)	Live	Dead	Log concentration	Mortality (%)	Probit unit	Prop (p)	Correct (p)	Logit (p)
20,000	2	0	20	0.30	100%		1	1	
10,000	1	0	20	0.00	100%		1	1	
5000	.5	0	20	-0.30	100%		1	1	
1000	.1	1	19	-1.00	95%	6.64	0.95	0.95	2.94
500	.05	3	17	-1.30	85%	6.04	0.85	0.85	1.73
250	.025	5	15	-1.60	75%	5.67	0.75	0.75	1.10
125	.0125	5	15	-1.90	75%	5.67	0.75	0.75	1.10
0	0	20	0		0%		0		

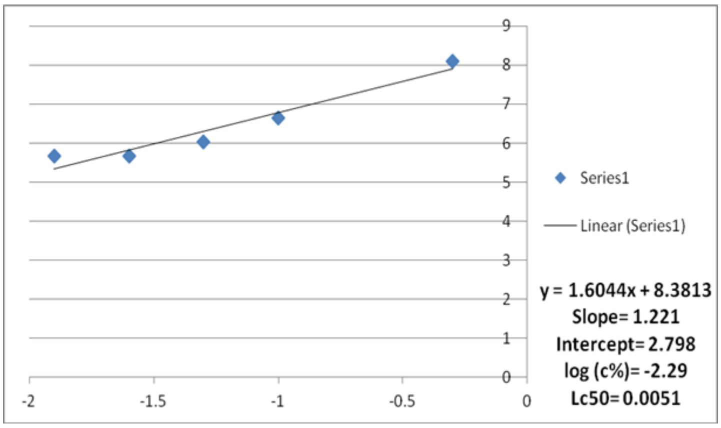

Figure 1. Trial 1

Table 2. Trial 2, Khartoum, 2017

Conc. PPM	Conc. (%)	Live	Dead	Log concentration on	Mortality (%)	Probit unit	Prop (p)	Correct (p)	Logit (p)
10000	1	0	20	0.00	100%		1	1	
5000	0.5	0	20	-0.30	100%		1	1	
1000	0 .1	3	17	-1.00	85%	6.04	0.85	0.85	1.73
500	0 .05	3	17	-1.30	85%	6.04	0.85	0.85	1.73
250	0 .025	5	15	-1.60	75%	5.67	0.75	0.75	1.10
125	0.0125	5	15	-1.90	75%	5.67	0.75	0.75	1.10
0		20	0				0	0	

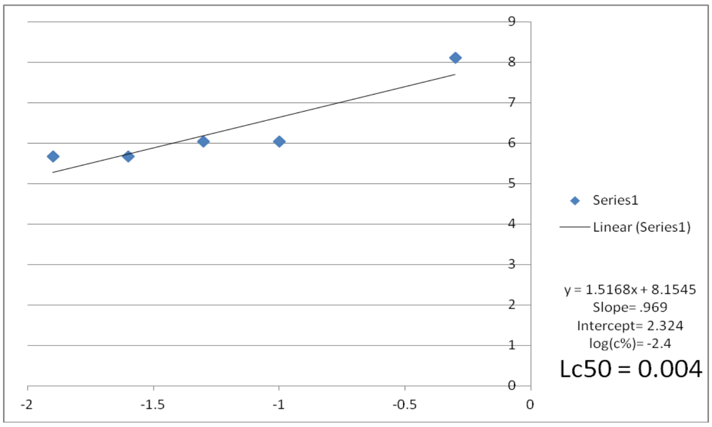

Figure 2. Trial 2

Table 3. Trial 3

Conc. PPM	Conc. (%)	Live	Dead	Log concentration on	Mortality (%)	Probit unit	Prop (p)	Correct (p)	Log it (p)
5000	0 .5	0	20	-0.30	100%		1	1	
1000	0.1	2	18	-1.00	90%	6.28	0.90	0.90	2.20
500	0.05	3	17	-1.30	85%	6.04	0.85	0.85	1.73
250	0.025	3	17	-1.60	85%	6.04	0.85	0.85	1.73
125	0.0125	6	14	-1.90	70%	5.52	0.70	0.70	0.85
0		20	0		0%		0		

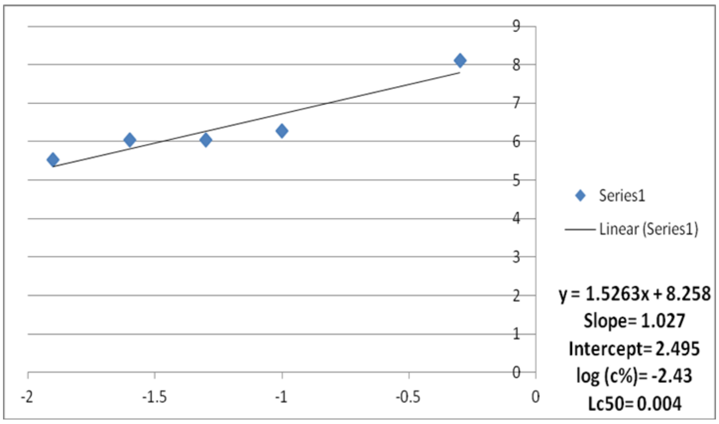

Figure 3. Trial 3

Table 4. Average of 3 trials, Khartoum, 2017

PPM	Conc. (%)	Log concentration	Prop (P)	Correct (p)	Logit (P)	Probit unit	Mortality (%)
20.000	2	0.30	1	1			100%
10.000	1	0.00	1	1			100%
5000	.5	-0.30	1	1			100%
1000	.1	-1.00	0.90	0.90	2.20	6.28	90%
500	0.05	-1.30	0.85	0.85	1.73	6.04	85%
250	0.025	-1.60	0.80	0.80	1.39	5.84	80%
125	0.0125	-1.90	0.75	0.75	1.30	5.67	75%
0	0		0				0%

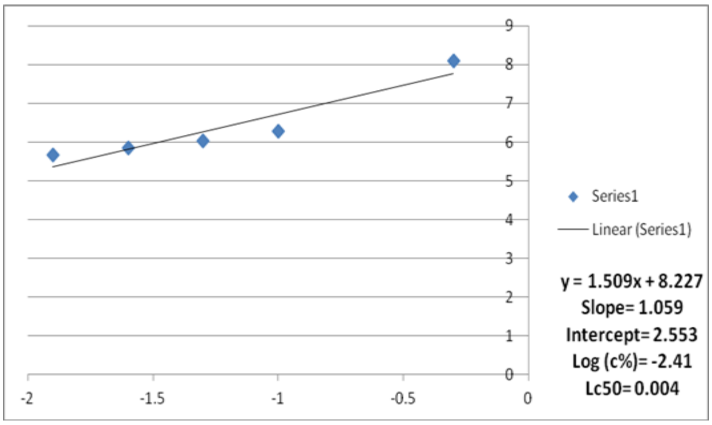

Figure 4. Average mortality of three trials

The products used for controlling larvae got attention, particularly those derived from plants. This study aimed to evaluate the efficacy of the aqueous extract of *Acacia nilotica* on mosquito larvae. While numerous studies in Sudan have explored other local plants, no published research has focused on *Acacia nilotica* for this purpose. This evergreen shrub is widespread across Sudan and can adapt to various soil types and climates. It is worth mentioning that soil composition may impact the concentration of active compounds in the plant. Additionally, extracts from different parts of the plant, such as seeds or bark, may yield different results. The LC_{50} values found in this study are comparable to those of some standard larvicides already in use.

While the entomological properties of *Acacia nilotica* remain underexplored, the results from this study are consistent with previous findings, such as those by Chaubal *et al*. [14], who observed that raw extracts from *Acacia nilotica*, particularly acetone extracts at a 500-ppm concentration, exhibited chronic toxicity to mosquito larvae like *Aedes aegypti* and *Culex quinquefasciatus*. This supports the idea that the active compounds in *Acacia nilotica* may play a significant role in controlling mosquito populations. However, extracting and standardizing these active ingredients are vital for ensuring consistent results and determining the plant's full potential as an insecticide.

To build on these findings, future studies should consider the impact of extracts from different parts of the plant and those derived from various soil types, as these factors influence the concentration and effectiveness of the active ingredients. In addition, expanding research to include solvents other than water may lead to more potent or versatile formulations. Given its environmentally friendly profile, cost-efficiency, and safety for non-target species, *Acacia nilotica* offers promising potential as a sustainable and safer alternative. This could reduce the health risks associated with the widespread use of synthetic pesticides.

Conclusion

The aqueous extract of *Acacia nilotica* has proven to be highly toxic to mosquito larvae, making it a promising candidate for larval control. Purifying the active compound responsible for this toxicity and investigating the effects of extracts from other solvents, as well as plant parts and soils, to further refine its efficacy. Continued research into the use of plant extracts that are environmentally safe, cost-effective, and non-hazardous to non-target organisms is vital, as these natural alternatives present a safer and more sustainable option compared to conventional insecticides.

Acknowledgments: None

Conflict of Interest: None

Financial Support: None

Ethics Statement: None

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