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Larvicidal Activity of Aqueous Extracts from *Jatropha curcas* on Mosquito Larvae from Kyambogo Swamp, Kyambogo University, Kampala-Uganda

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Key words: Larvicidal activity, *Jatropha curcas*, Mosquito larvae

Abstract: Mosquito transmitted infections are the leading cause of morbidity and mortality in many African countries including Uganda. The mosquitoes lay their eggs in stagnant water. The eggs then hatch into larvae, pupa and later into adults. Synthetic insecticides are commonly used to control these deadly insects. However, most of these insecticides are very expensive and non-environmentally friendly. There is also increased resistance of mosquitoes to these insecticides hence the need to search for new sources of bioactive compounds that can potentially deplete and incapacitate mosquito populations. *Jatropha curcas* has been reported to contain larvicidal, insecticidal and insect repellency properties. This study therefore aimed at assessing the larvicidal activity of aqueous extracts from *Jatropha curcas* seeds, leaves, barks and roots on mosquito larvae in vitro using a Completely Randomized Experimental Design. The seed extract exhibited a mortality rate of 83.3% at 80% concentration level and 80, 60, 56.7 and 40% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. The leaf extract exhibited a mortality rate of 43.3% at 80% concentration level and 33.3, 23.3, 13.3 and 10% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. The bark extract exhibited a mortality rate of 43.3% at 80% concentration level and 26.7, 20, 13.3 and 6.7% mortality rates at 60, 40, 20 and 10% concentration levels, respectively while the root extract exhibited a mortality rate of 43.3% at 80% concentration level and 23.3, 20, 13.3 and 6.7% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. In the control experiments, no mortality of mosquito larvae was noted. All the aqueous *Jatropha curcas* extracts were able to cause mortality of mosquito larvae within a period of 24 h. The mortality rates generally increased with increase in concentration levels of the extracts. However, apart from the bark extract, increase in the concentration levels of the rest

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of the extracts did not yield any significant difference in the mortality rates of the mosquito larvae at 5% significance level. Generally, the seed extract

exhibited the highest larvicidal activity hence it could be used in the development of affordable, effective and environmentally friendly mosquito larvicides.

INTRODUCTION

Mosquito transmitted infections, especially malaria are the leading cause of morbidity and mortality in many African countries including Uganda hence a very big public health threat^[1]. In fact, Uganda has the 3rd highest global burden of malaria cases (5%) and the 7th highest level of deaths^[1]. It also has the highest proportion of malaria cases in East and Southern Africa (23.7%)^[1]. Malaria is caused by *Plasmodium* spp. which are transmitted by infected female the anopheles mosquitoes. The mosquitoes lay their eggs in stagnant water which hatch into larvae and later into adults. Various methods have been used in controlling mosquitoes and their larvae with the use of insecticides being the commonest method. Insecticides are usually applied directly on the mosquito breeding or resting sites (indoor and outdoor spray) to kill the larvae and adults^[2]. However, most of the available insecticides on the market are very expensive and toxic to even non target organisms^[2]. Furthermore, there is increased resistance of mosquitoes to the available insecticides^[2]. The second method involves elimination or manipulation of mosquito habitats and breeding sites such as containers, tyres and drums where water can stagnate to stop mosquitoes from laying their eggs^[3]. However, this method is very tedious, time consuming and less effective. Alteration of the mosquito breeding environment to make it unfavorable for mosquitoes to breed is another method^[3]. However, this method is very tedious, time consuming and it involves technical knowhow ecology and biology of mosquitoes. The use of parasites, predators, pathogens and symbionts of mosquitoes is another method that has been used in biologically controlling mosquitoes and their larvae. The most successful bio-control agents to date are fish and the bacteria known as *Bacillus thuringiensis israelensis* (*Bti*) and *Bacillus sphaericus* (*Bs*). Much as this method is environmentally friendly and there is much less risk of mosquitoes developing resistance, some of the bio-control agents are less specific and process of identifying an effective bio-control agent is very expensive and tedious^[4]. Personal protection which entails persons protecting themselves from mosquito bites using repellent, mosquito coils, nets or clothing is another common control method^[4]. However, most of the materials used for personal protection against mosquito bites are very expensive and not readily available, especially in the rural areas. This has created the need to search for new effective, cheap, environmentally friendly and readily

available methods that can be used to deplete and incapacitate mosquito populations probably by use of plantbased materials. Globally, different plant extracts have been used to control mosquito larvae. For instance, *Artemisia annua*, *Acacia nilotica*, *Argemone mexicana* leaf and seed extracts have been previously used as larvicides. *Jatropha curcas* leaf and root extracts, *Withania somnifera*, *Citrullus colocynthis* and *Cannabis sativa* leaf extracts have also been previously used in controlling adult mosquitoes^[5]. *Jatropha curcas* has also been reported to contain insecticidal properties^[6]. This study therefore aimed at assessing the larvicidal activity of aqueous extracts from *Jatropha curcas* seeds, leaves, bark and roots on mosquito larvae *in vitro*.

MATERIALS AND METHODS

Collection of the plant parts: *Jatropha curcas* seeds, leaves, bark and roots were collected from Kyambogo University Farm, Nakawa Division, Kampala, Uganda. The collected plant parts were packed in separate plastic containers as described by Gitonga *et al.*^[7]. The containers were then labeled and transported to the Biology Research Laboratory of Kyambogo University.

Processing of the plant parts: In the laboratory, the collected plant parts were sorted and separately air dried in a well-ventilated place for a period of one week (7 days) to reduce their moisture content. The plant parts were then separately pounded using a mortar and pestle and the resulting powder sieved through a 0.5 mm pore size mesh to obtain uniform fine powdery particles. This was followed by weighing 10, 20, 40, 60 and 80 g of the powdery particles from each of the plant parts using an analytical weighing balance with an error correction of ± 0.001 . Each weighed quantity of the powder was separately added to a clearly labeled conical flask containing 100 mL of distilled water. The contents in each flask were thoroughly stirred using a magnetic stirrer to evenly distribute the powder. The flasks were plugged with cotton wool and left to stand at room temperature for a period of 48 h. The contents in each flask were then filtered using a wire strainer followed by Whatman number 1 filter paper to obtain filtrates of 10, 20, 40, 60 and 80% (w/v) concentration levels for each plant part.

Collection of mosquito larvae: The mosquito larvae were collected from a pond in Kyambogo Swamp that is located in Nakawa Division, Kampala-Uganda. The water

containing the mosquito larvae was carefully scooped using a bucket and immediately transported to Kyambogo University Biology Laboratory for the bioassays. In the laboratory, the water was left for 30 min to allow the larvae settle at the bottom of the bucket. The water was then reduced by decanting to enable easy counting and scooping of the larvae.

Bioassay procedure for the larvicidal activity: Using a Completely Randomized Experimental Design (72) clean plastic petri dishes were arranged on a clean laboratory bench. The 20 milliliters of the different concentration levels (10, 20, 40, 60 and 80%) of each extract separately added to the petri dishes in triplicates with distilled water acting as the control. To each of the petri dishes containing the extracts, (10) live mosquito larvae were introduced and the experiment left to run for 24 h. After the 24 h, dead larvae in each petri dish were counted, recorded and the percentage mortality at each concentration level of the plant part extract calculated using the formula below prior to summarizing the results in form of average mortality rates. Percentage mortality = $\frac{\text{Number of dead larvae} \times 100}{\text{sum of larvae in a petri dish}}$.

RESULTS AND DISCUSSION

Larvicidal activity of *J. curcas* seed extract: All the concentration levels (10, 20, 40, 60 and 80%) of *Jatropha curcas* seed extract exhibited larvicidal potency against mosquito larvae. The seed extract exhibited a mortality rate of 83.3% at 80% concentration level and 80, 60, 56.7 and 40% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. However, $p > 0.05$ and $F_{crit} (4.458) > F (0.78)$, hence, there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* seed extract (Table 1). The larvicidal activity of the *J. curcas* seed extract could be attributed to the presence of cursin, oleic and linoleic acids which are known to have insecticidal activities^[8]. Abdelgadir and Van Staden^[9] attributed the larvicidal activity of the seed extract to the presence of other toxic chemical compounds such as saccharose, raffinose, stachyose, glucose, fructose, galactose and toxic proteins. In a study that was carried out by Rahuman *et al.*^[10] using seed oil of *J. curcas* against

Anopheles mosquito larvae at 10-50% concentration, 50-86% mortality rates within 24 h was registered. This is in agreement with the results from the present study.

Larvicidal activity of *J. curcas* leaf extract: All the concentration levels (10, 20, 40, 60 and 80%) of *Jatropha curcas* leaf extract exhibited larvicidal potency against mosquito larvae. The leaf extract exhibited a mortality rate of 43.3% at 80% concentration level and 33.3, 23.3, 13.3 and 10% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. However, $p > 0.05$ and $F_{crit} (4.458) > F (2.154)$, hence, there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* leaf extract (Table 2). The larvicidal activity of *J. curcas* leaf extract could be attributed to the presence of bioactive compounds like glycosides, tannins, phytosterols, flavonoids and sterols, saponins in *J. curcas* that have larvicidal effects on *Anopheles* mosquito larvae^[11]. The larvicidal activity could also be due to the presence of flavones like apigenin, orientin and vitexin in *J. curcas* leaves^[12]. Tomass *et al.*^[13] had earlier reported that crude and column chromatographic fractions of the methanol leaf extracts of *J. curcas* have larvicidal activities against the laboratory-reared late third instar larvae of *Anopheles* mosquito larvae. It elicited larvicidal activity ranging from 12.5-50% at concentration levels of 10-50% within 24 h, hence, results being similar to 10-43.3% mortality rate at 10-80% concentration levels, respectively registered in the present study.

Larvicidal activity of *J. curcas* bark extract: All the concentration levels (10, 20, 40, 60 and 80%) of *Jatropha curcas* bark extract exhibited larvicidal potency against mosquito larvae. The bark extract exhibited a mortality rate of 43.3% at 80% concentration level and 26.7, 20, 13.3 and 6.7% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. However, $p < 0.05$ and $F_{crit} (4.458) < F (6.0)$, hence, there was a significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* bark extract (Table 3). *Jatropha curcas* bark stores a vast number of phytochemical compounds that have biological action on

Table 1: Larvicidal activity of *J. curcas* seed extract

Concentration level (%)	80	60	40	20	10
Average mortality rate (%)	83.3	80	60	56.7	40
$p = (0.489)$	$F (0.78)$	$F_{crit} (4.458)$			

$p > 0.05$ and $F_{crit} (4.458) > F (0.78)$, hence there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* seed extract

Table 2: Larvicidal activity of *J. curcas* leaf extract

Concentration level (%)	80	60	40	20	10
Average mortality rate (%)	43.3	33.3	23.3	13.3	100
$p = (0.178)$	$F (2.154)$			$F_{crit} (4.458)$	

$p > 0.05$ and $F_{crit} (4.458) > F (2.154)$, hence, there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* leaf extract

Table 3: Larvicidal activity of *J. curcas* bark extract

Concentration level (%)	80	60	40	20	10
Average mortality rate (%)	43.3	26.7	20	13.3	6.7
p = (0.0256)	F (6.0)		Fcrit (4.458)		

$p < 0.05$ and $F_{crit} (4.458) < F (6.0)$, hence, there was a significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* bark extract

Table 4: Larvicidal activity of *J. curcas* root extract

Concentration level (%)	80	60	40	20	10
Average mortality rate (%)	43.3	23.3	20	13.3	6.7
p = (0.8145)	F (0.2105)		Fcrit (4.458)		

$p > 0.05$ and $F_{crit} (4.458) > F (0.2105)$, hence, there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* root extract

mosquito larvae. These compounds include; alkaloids, saponins and tannins^[14]. In the present study, the larval mortality rate increased with increase in concentration levels of the extract as evidenced by Thongwat *et al.*^[15]. The presence of steroids and ethanol in *J. curcas* could be another cause of the larval mortality^[15]. Gunasekaram *et al.*^[16] used undiluted *J. curcas* bark crushed in a motor into watery paste and registered 48-98% larval mortality at concentration levels between 5-30% in a period of 12 h. The results (10-43.3% mortality at 10-80% concentration level within 24 h) in the present study do not agree with those of [16] where an undiluted bark extract was used. Therefore, the concentration of phytochemicals in the undiluted bark extract could have been higher than the one used in present study which entailed dilutions. Furthermore, some phytochemicals could have evaporated during the process of drying and pounding the bark leading to reduced larvicidal activity.

Larvicidal activity of *J. curcas* root extract: All the concentration levels (10, 20, 40, 60 and 80%) of *Jatropha curcas* root extract exhibited larvicidal potency against mosquito larvae. The root extract exhibited a mortality rate of 43.3% at 80% concentration level and 23.3, 20, 13.3 and 6.7% mortality rates at 60, 40, 20 and 10% concentration levels, respectively. However, $p > 0.05$ and $F_{crit} (4.458) > F (0.2105)$, hence, there was no significant difference in the mortality rates of the mosquito larvae at the different concentration levels of *J. curcas* root extract (Table 4). Experimentation of methanol root extract of *J. curcas* against *Anopheles* mosquitoes did not show significant larvicidal activity^[17]. Only 20-40% mortality was observed up to 10-50% concentration of extract within 24 h, hence, this is in agreement with 3.3-40% mortality at 10-80% concentration levels registered in the present study within the same time frame (Table 4).

Apart from the bark extract, increase in the concentration levels of *J. curcas* seed, leaf and root extracts did not cause any significant change in larvicidal activity.

CONCLUSION

All the aqueous *Jatropha curcas* extracts were able cause mortality of mosquito larvae within a period of 24 h. The mortality rates generally increased with increase in concentration levels of the extracts. However, apart from the bark extract, increase in the concentration levels of the rest of the extracts did not yield any significant difference in the mortality rates of the mosquito larvae at 5% significance level. Generally, the seed extract exhibited the highest larvicidal activity hence it could be used in the development of affordable, effective and environmentally friendly mosquito larvicides.

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