

# Larvicidal Activity of *Artocarpus Altilis* against *Culex Quinquefasciatus*

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## Abstract

*Artocarpus altilis* is an evergreen, flowering tree in the family Moraceae. Its parts have antimalaria and insect repellent activities. The methanol extract of the stem bark and the wood was reported to have moderate to low activity respectively against the fourth instar larvae of *Aedes aegypti*. In this study, the methanol extract of the leaf, root, stem bark, root bark, flower, fruit and wood of *A. altilis* was tested against the fourth instar larvae of *Culex quinquefasciatus* mosquito. This was with a view to determining the most active morphological part from which eco-friendly and potent larvicidal compound(s) could be isolated. The flower ( $LC_{50}$  2.19  $\pm$  0.16mg/mL at 48h) and the root ( $LC_{50}$  2.18  $\pm$  0.09mg/mL at 48h) extracts had the highest larvicidal activity. The root extract was partitioned into n-hexane and ethylacetate and the resulting fractions tested. The ethylacetate fraction ( $LC_{50}$  1.01  $\pm$  0.03mg/mL at 48h) was the most active.

**Keywords:** *Artocarpus altilis*; partitioned fraction; filariasis; *Culex quinquefasciatus*; Moraceae; larvicidal activity;

## Introduction

*Artocarpus altilis* commonly known as breadfruit is an evergreen, flowering tree in the family Moraceae [1]. The leaves are thick and leathery with a glossy dark-green colour on the dorsal side. The underside is dull with an elevated midrib and main veins [2]. The wood is gold in colour, resistant to termites and shipworms, so it is used as timber for structures and outrigger canoes. Breadfruit tree bears a multitude of monoexious flowers. The fruits are mostly round, oval, or oblong in shape. The seeds are brown in colour, shiny, round or ovoid in shape and irregularly compressed. It is a multipurpose agroforestry tree crop which is primarily used for its nutritious, starchy fruit as rich source of carbohydrates, calcium and phosphorus [3]. The different parts are used for the treatment of tongue thrush, skin infections, sciatica, diarrhoea, low blood pressure and asthma [4, 5]. It has been reported as having potential as an insect repellent [6]. Anti-inflammatory, antifungal, antidiabetic, immunomodulatory, antitubercular, antiplasmodial, antihypertensive, antibacterial, anti-cholinergic, chelating, toxicity to cancer cells and anthelmintic activities had been reported for the plant [7-14]. Many compounds like morin, moracin, dihydromorin, cynomacurin, cyclomorusin,

artocarpin, artocarpetin, cycloartinone, cyclogeracommunin, and cyclocommunol and cycloartenyl acetate had been isolated from the plant [15, 16]. The methanol extract of the fruit showed poor insecticidal and larvicidal activity against *Bruchus pisorum*, *Tribolium castaneum* and *Sitophilus oryzae* [17]. The methanol extracts of the stem bark and wood was reported to have moderate to low larvicidal activity respectively against *A. aegypti* [18]. Fatty acids were suggested to be responsible for the mosquito deterrence of the hydrodistillate of the dried male inflorescences against adult *A. aegypti* females [19]. This paper reports the larvicidal activity of the various morphological parts of *A. altilis* against *Culex quinquefasciatus* the vector of filariasis. This was with a view to determining the most active morphological part from which eco-friendly and potent larvicidal compound(s) could be isolated.

## Methods

### Plant Collection and Preparation

The leaves, root, stem bark, root bark, flowers, fruits and wood of *A. altilis* (Parkinson) Forsberg (Moraceae) were collected on a farm land along link road, Obafemi Awolowo University Hospital, Ile-Ife, Osun state. The plant was authenticated by Mr Ogunlowo of the Herbarium of the Department of Pharmacognosy, Obafemi Awolowo University, Ile-Ife, Osun state. Voucher specimen was deposited under the reference number FPI 2177. The collected morphological parts were cut into smaller pieces. The leaf and flower were air dried while the root, root bark, wood and stem bark were oven dried at 40°C. They were separately blended in a grinding machine. The fresh fruit was pounded in a mortar with a pestle. Each of the plant parts was extracted in methanol at room temperature for 3 days, with agitation. The extract was filtered and concentrated *in vacuo* at 35°C. This was repeated twice. The combined extract for each plant part was kept and later used for larvicidal activity testing against *C. quinquefasciatus*.

### Larvicidal Activity of the Extracts

Each of the resulting extracts was subjected to larvicidal activity testing according to World Health Organisation, 2005 guidelines with slight modifications. Stock solutions (25 mg/mL)

of the extracts prepared by solubilising the extracts in Tween 80 were thereafter serially diluted to obtain 25 mL of different concentrations (0–5 mg/mL) of the test agents. Twenty five larvae were introduced into each cup and each concentration was replicated five times. The negative control contained distilled water and Tween 80 and Endosulphan, a commercial insecticide, was used as the positive control. The number of surviving larvae in each cup was counted after 24 and 48 hours of exposure. Average percentage mortality for each concentration was calculated from which the  $LC_{50}$  and  $LC_{90}$  values were determined [18]. No mortality was observed with the negative control.

### Partitioning of the Root Extract

The methanol extract of the root (AAR, 9.48 g) was suspended in water and successively partitioned into n-hexane and ethylacetate; and concentrated *in vacuo* to give their corresponding n-hexane (AAR1, 1.2 g), ethylacetate (AAR2, 1.5 g) and aqueous (AAR3, 6.38g) fractions.

### Larvicidal Activity of the Partitioned Fractions

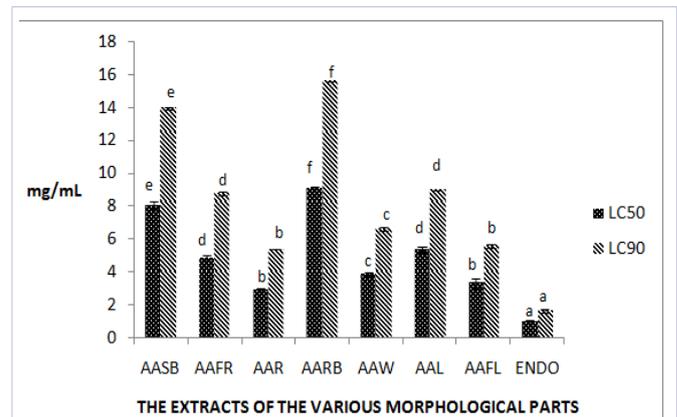
Stock solutions (12.5 mg/mL) of each of the partitioned fractions AAR1-AAR3 prepared by solubilising them in Tween 80 were thereafter serially diluted to obtain 25 mL of different concentrations (0–2.5 mg/mL) of the test agents. These were used for the assay as given for the extracts.

## Results

Presented in figures

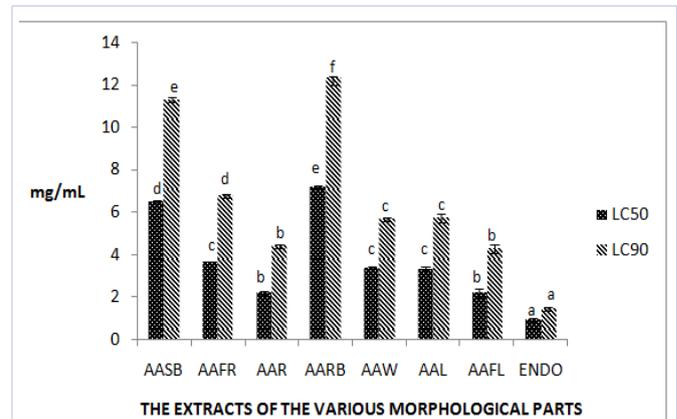
## Discussion

The various morphological parts of *A. altilis* were screened for activity against the fourth instar larvae of *C. quinquefasciatus* the vector of filariasis and many other debilitating diseases. During the test period, the methanol extract of the various parts demonstrated varying degrees of activity against the test organism. At both 24 and 48 h, the root and flower extracts had the highest activity (Figure 1 and 2). The high activity of the flower corroborated an earlier report of mosquito deterrence of the hydrodistillate of the dried male inflorescences against adult *A. aegypti* females [19]. The leaf, wood and fruit extracts were moderately active while the stem and root barks were inactive as shown in Figs. 1 and 2 [20]. However none of the extracts had comparable activity to the positive control used. The order of activity was Endo > root = flower > wood > leaf = fruit > stem bark > root bark at 24hours. At 48h, there was a significant improvement in activity of all the extracts most especially the fruit and leaf. The order of activity was Endo > root = flower > wood = leaf = fruit > stem bark > root bark. The activity of the wood extract ( $LC_{50}$ ,  $LC_{90}$  3.83 ± 0.08, 6.6 ± 0.07 mg/mL at 24 hours) of this study against *C. quinquefasciatus* was significantly better than that reported against *A. aegypti* ( $LC_{50}$ ,  $LC_{90}$  6.38 ± 0.29, 10.33 ± 0.22 mg/mL at 24 hours) by Adebajo et al., 2014. This could be due to a higher susceptibility of the test organism used in this study.



**Figure 1:** The Larvicidal Activity of the Methanol Extract of the Various Morphological Parts after 24 hours.

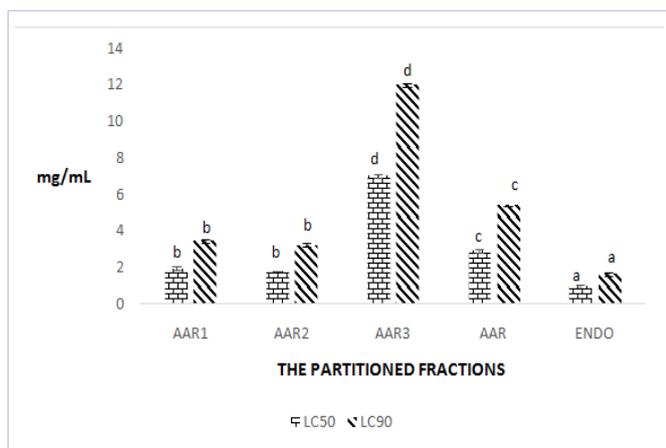
AASB: stem bark extract; AAFR: fruit extract; AAR: root extract; AARB: root bark extract; AAW: wood extract; AAL: leaf extract; AAFL: flower extract; ENDO: Endosulphan, the positive control.  $LC_{50}$  and  $LC_{90}$ : Values ± SEM of five experiments.



**Figure 2:** The Larvicidal Activity of the Methanol Extract of the Various Morphological Parts after 48 hours.

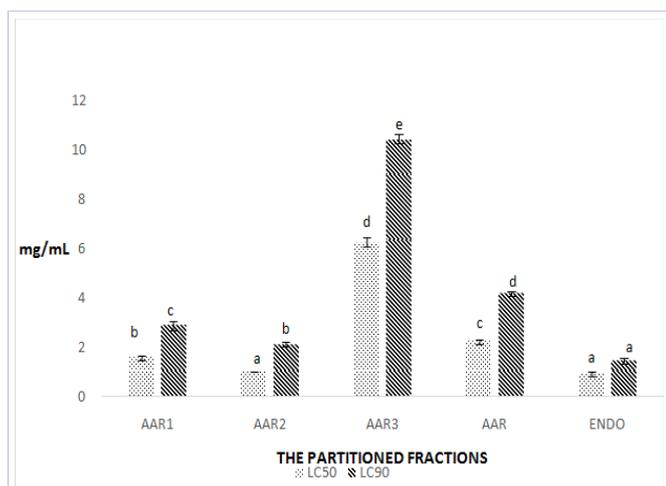
AASB: stem bark extract; AAFR: fruit extract; AAR: root extract; AARB: root bark extract; AAW: wood extract; AAL: leaf extract; AAFL: flower extract; ENDO: Endosulphan, the positive control.  $LC_{50}$  and  $LC_{90}$ : Values ± SEM of five experiments.

The methanol extracts of the root and flower gave comparably highest activity. However, the yield of the methanol extract of the flower was low compared to the root. Therefore in order to have a good weight for phytochemical work, the root was further purified. The methanol extract of the root was suspended in water and successively partitioned into n-hexane and ethylacetate. The resulting n-hexane, ethylacetate and aqueous fractions were similarly tested against the fourth instar larvae of *C. quinquefasciatus*. At 24 hours, the n-hexane ( $LC_{50}$ ,  $LC_{90}$  1.92 ± 0.08, 3.41 ± 0.12 mg/mL) and the ethylacetate ( $LC_{50}$ ,  $LC_{90}$  1.77 ± 0.03, 3.19 ± 0.13 mg/mL) partitioned fractions had comparable high larvicidal activity (Figure 3 and 4) while the aqueous fraction was inactive ( $LC_{50}$ ,  $LC_{90}$  6.99 ± 0.12, 11.97 ± 0.13 mg/mL). The activities of the organic fractions were better than that of the methanol extract ( $LC_{50}$ ,  $LC_{90}$  2.87 ± 0.08, 5.37 ± 0.01 mg/mL). At



**Figure 3:** The Larvicidal Activity of the Partitioned Fractions of the Root Extract after 24 hours.

**Key:** **AAR1:** n-hexane partitioned fraction; **AAR2:** ethylacetate partitioned fraction; **AAR3:** aqueous partitioned fraction; **AAR:** root extract; **ENDO:** Endosulphan, the positive control. LC<sub>50</sub> and LC<sub>90</sub>: Values ± SEM of five experiments.



**Figure 4:** The Larvicidal Activity of the Partitioned Fractions of the Root Extract after 48 hours.

**Key:** **AAR1:** n-hexane partitioned fraction; **AAR2:** ethylacetate partitioned fraction; **AAR3:** aqueous partitioned fraction; **AAR:** root extract; **ENDO:** Endosulphan, the positive control. LC<sub>50</sub> and LC<sub>90</sub>: Values ± SEM of five experiments

48 hours, the ethylacetate fraction had the lowest lethality value (LC<sub>50</sub>, LC<sub>90</sub> 1.01 ± 0.03, 2.02 ± 0.06 mg/mL) making it the most active partitioned fraction. Its activity was comparable to that of the positive control. Efforts are on-going to isolate the active compound(s) from the most active ethylacetate fraction.

## Conclusion

The results obtained from this study indicate that *A. altilis* root and flower extracts could serve as potential candidates for developing botanical larvicides for efficient control of *C. quinquefasciatus*. The larvicidal compound(s) of the root extract is concentrated in the ethylacetate fraction and efforts are on-going to isolate them.

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