

## Damage caused by castor seed oil and its ethanol extracts on *Anopheles arabiensis* Larvae, Gezira State, Sudan

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### Abstract

*Anopheles* mosquitoes are considered as vector of malaria disease and some other endemic diseases in the world. There are some methods already been used for controlling mosquito; of which is natural products. This study was conducted at Laboratories of Faculty of Engineering and Technology, University of Gezira, to evaluate the damage caused by Castor seed raw-oil and its ethanol extract on *Anopheles arabiensis* mosquito larvae. The Castor seeds were collected from Wad Medani City, whereas, the mosquito larvae were collected from the breeding sites at Tayba village, Gezira State, Sudan. The Castor seeds were shade dried away from the direct sunlight, grounded and then kept separately in small plastic sacks. The raw oil (extracted from the castor seeds) was applied at a concentration of 1.67 ml/L (0.50 ml oil/ 300 ml water) while its ethanol extract was applied at concentration of 30.53 mg/L. The standards of WHO for testing the toxic compound against mosquito larvae was followed. The mortality in *A. arabiensis* larvae were 95% and 100%, respectively, for the ethanol extract and raw oil, after 24 hours. The results also showed the damage caused by each treatment on the dead larvae and on the survived larvae. The damage involved changes in skin color, formation of larval-pupal intermediate form, swelling, separation in digestive tract, separation of body hairs and decapitation. The study recommends adding these castor seeds as potential natural products for *Anopheles* larval control, and also running more sensitive tests on human, environmental resources and on the aquatic fauna.

**Keywords:** Castor, Seed Oil, Ethanol Extracts, *Anopheles arabiensis*, Larvae

### 1. Introduction

Mosquitoes are important vectors of several tropical diseases. About one hundred species act as vectors of human diseases [6] and some species were reported to transmit arboviruses [16]. Other biting members of *Anopheles* spp are nuisance to man [7]. The mean number of *A. arabiensis* larvae collected from Wad Medani, Gezira State, was significantly higher than the *Anopheles* species [1]. Mosquitoes can transmit more diseases pathogens than any other group of arthropods and affect millions of people throughout the world. WHO has declared the mosquitoes as "public enemy number one". Mosquito borne diseases are prevalent in more than 100 countries across the world, infecting over 700 million people every year globally and 40 million of the Indian population. They act as a vector for most of the life threatening diseases like malaria, yellow fever, dengue fever, chikungunya fever, filariasis, encephalitis, West Nile virus infection, etc., in almost all tropical and subtropical countries and many other parts of the world [17]. The major tool in mosquito control operation is the application of synthetic insecticides such as organochlorine and organophosphate compounds. But this has not been very successful due to human, technical, operational, ecological, and economic factors. In recent years, use of many of the former synthetic insecticides in mosquito control program has been limited. It is due to lack of novel insecticides, high cost of synthetic insecticides, concern for environmental sustainability, harmful effect on human health, and other non-target populations, their non-biodegradable nature, higher rate of biological magnification through ecosystem, and increasing

insecticide resistance on a global scale [5, 13]. Thus, the Environmental Protection Act (EPA) has framed a number of rules and regulations to check the application of chemical control agents in nature [4]. Within the field of organic chemistry, the definition of natural products is usually restricted to mean purified organic compounds isolated from natural sources that are produced by the pathways of primary or secondary metabolism. Within the field of medicinal chemistry, the definition is often further restricted to secondary metabolites [17]. Secondary metabolites are not essential for survival, but nevertheless provide organisms that produce them an evolutionary advantage [10]. Many secondary metabolites are selected and optimized through evolution for use as "chemical warfare" agents against prey, predators, and competing organisms [3].

They are 1,200 plant species having potential insecticidal value [12] the listed and discussed 344 plant species that only exhibited mosquitocidal activity [15]. The reviewed the state of knowledge on larvicidal plant species, extraction processes, growth and reproduction inhibiting phytochemicals, botanical ovicides, synergistic, additive and antagonistic joint action effects of mixtures, residual capacity, effects on non-target organisms, resistance and screening methodologies, and discussed some promising advances made in phytochemical research [14].

Castor (*Ricinus communis*) is green shrub with large palmate leaves and apical racemose flowers. Castor oil composed of glycerides of several fatty acids, whereas ricinoleic acid is of most important. The oil contains also the ricinine alkaloid. The

seeds contains a very toxic resin <sup>[2]</sup>. *R. communis* known to either stop the cell division proliferating tissues resulting in sterility in insects or disrupt their physiological functions <sup>[11]</sup>. Seed oil contains a very toxic lectin, ricin and ricinine <sup>[9]</sup>. Another possible and quite serious side effect of castor oil is the swelling of the face, lips, tongue or throat. In such cases, immediate medical intervention is absolutely recommended <sup>[8]</sup>. This study aimed to assess the damage caused by the castor (*Ricinus communis*) as raw oil and also its ethanol extract on the mosquito (*Anopheles arabiensis*) larvae.

## 2. Materials and Methods

### 2.1 The Study Area

Wad Medani City is located in the central parts of the Gezira State, Sudan. Tayba village was selected for sampling mosquitoes (North Wad Medani City), whereas, Castor seeds were collected from within Wad Medani City.

### 2.2 Methodology

Larvae of *A. arabiensis* were collected with sufficient amounts of breeding water. Rearing and maintenance of mosquito larvae followed <sup>[17]</sup>. The collected castor seeds were shade dried at room temperature. The dried seeds were grounded, using mortar and pestle and were then used for extracting raw oil (and its ethanol extract).

Experiments were started by preparing sufficient number of plastic cups (size of about 350 ml). Random samples (20 individuals) of *A. arabiensis* larvae of the third or early fourth instar were placed on these cups which were adjusted to 300 ml tap water. About 0.5 ml of raw oil was added (in the first test), and 0.5 ml of the ethanol extract (in the second test) to these cups that containing *A. arabiensis* larvae and 300 ml water. This experiment was triplicate. These experiments were run in the Laboratories of Faculty of Engineering and Technology, at the room temperature ( $26 \pm 3^\circ\text{C}$ ). After 24 hours, in each test cup, the dead larvae were counted, put in glass slides to be photographed for monitoring the morphological changes (Specially changes in color, status of digestive tract and the separation of some body parts. A digital microscope provided with camera was used for documentation of these observed changes.

### 2.3 Data Analysis

Data from the study experiment were collected and presented as Plates to reflect the damage observed in *Anopheles* larvae, so no statistical analysis was needed.

## 3. Results and Discussion

### 3.1 The damage caused by ethanol extract of castor seed-oil on *A. arabiensis* larvae

After 24 hours of applying ethanol extract of castor seeds (at 30.53 mg/L), and by using digital microscope provided with camera, lethal effect (Table, 1), some morphological changes were monitored on the larvae (Fig, 1). The mean percentage mortality in *A. arabiensis* larvae was 98.33% (only one larva out of 60 was survived). The lethal effect can be due to composition of seed oil which contains a very toxic lectin, ricin and ricinine <sup>[8]</sup>.

The change in the larval color (to brighter color) was noticed in most (95%) larvae subjected to the ethanol extract. There were few number of larvae with disconnected digestive tract (about 10%). The reason may be referred to the fact that, ethanol may extract some potent chemicals from the raw oil, and these materials caused damage in the connective and other tissues of the digestive tracts of the tested larvae. *R. communis* known to stop the cell division proliferating tissues in insects and disrupt their physiological functions <sup>[11]</sup>.

About half (50%) of the larvae were decapitate as a result of being submitted to the ethanol extract of the castor raw oil, and this may be due to the effect of the materials being extracted from the castor oil that led to loosening the tissues connecting the head to the thorax. All (100%) the larvae were noticed to be swelled (not like the control larvae). The swelling effect caused by castor oil is widely observed <sup>[8]</sup>. Only one larva (1.67%) was failed to pupate, and an intermediate larva-pupa form was noticed <sup>[11]</sup>.

### 3.2 The damage caused by castor raw oil on *A. arabiensis* larvae

After 24 hour of applying castor seed-oil, a lethal effect and some morphological changes (in color, loss of the body hairs, swelling, decapitate and with sealed feeding brushes) in addition to failure to pupate on larvae of *A. arabiensis* were noticed (Table, 1 and Fig, 2).

The change in the larval color submitted to castor seed-oil was high (85%), and only (15%) retaining their original color. The castor seed oil resulted in mean mortality on the larvae of *A. arabiensis* of 100%. It was also noticed that, some of the body parts of the larvae were separated (head (decapitate in 45%) and body hairs (in about 65%)), also some of the larvae were obviously swollen (in about 80% of the larvae) in comparison to the control. Oldest larvae failed to pupate (noticed in only one larva; 1.67%) and some larvae sealed their feeding brushes (in about 55% of the larvae), and this phenomenon could be attributed to that, larvae used their feeding brushes to close their mouth openings to prevent swallowing of castor oil during breathing at water surface.

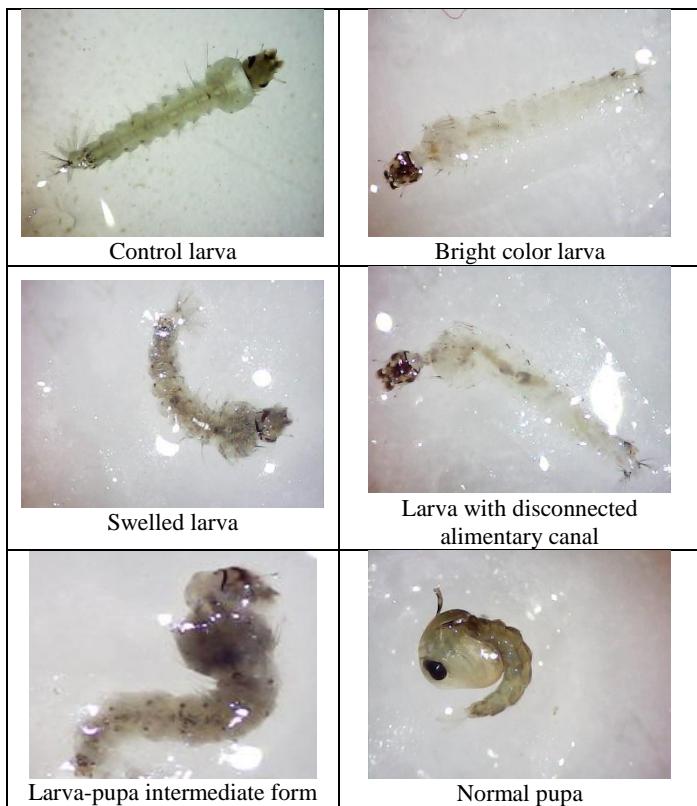
## 4. Conclusions

The ethanol extract of castor seed-oil (at 30.53 mg/L), and after 24 hours, caused lethal effect, loss in color (to brighter color), disconnecting digestive tract, decapitating, swelling and preventing pupation in *A. arabiensis* larvae. The raw castor seed-oil, after 24 hours, causing lethal effect and loss in color, loss of the body hairs, swelling, decapitate and sealed feeding brushes, on larvae of *A. arabiensis*.

The study recommends running more sensitivity tests of castor seed-oil on human, environmental resources and on the aquatic predators.

**Table 1:** Mortality (%) in *Anopheles arabiensis* larvae submitted to castor seed-oil (at 1.67 ml/L) and its ethanol extract (at 30.53 mg/L)

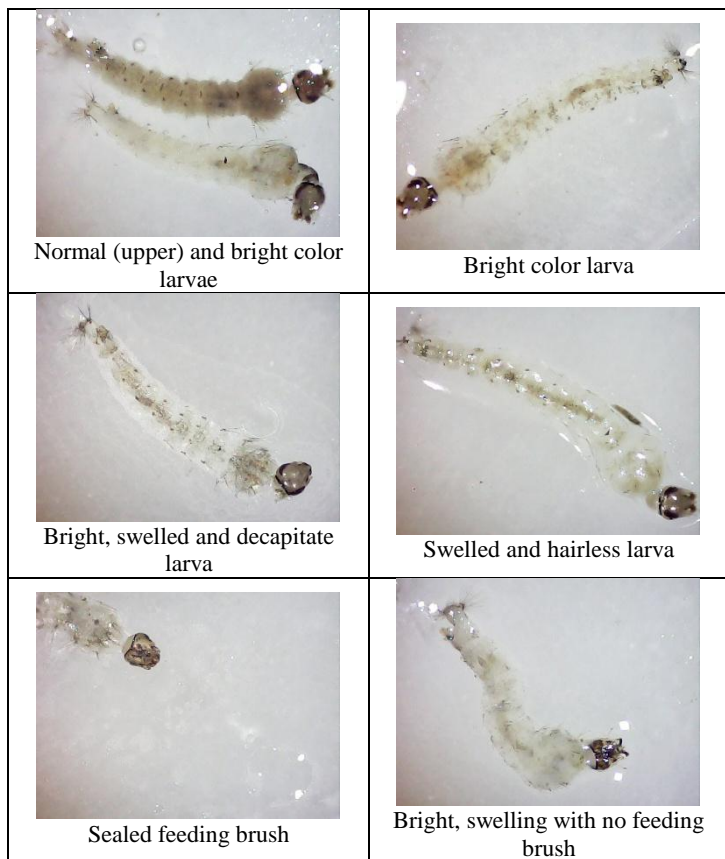
Rep.	Castor seed-oil (CSO)	Ethanol extract of CSO
1	100	100
2	100	100
3	100	95%
Mean	100	98.33



**Fig 1:** The damage caused by the ethanol extract of castor seeds (at 30.53 mg/L) on *Anopheles arabiensis* larvae after 24 hour

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**Fig 2:** The damage caused by castor seeds raw oil (at 1.67 ml/L) on *Anopheles arabiensis* larvae after 24 hour