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Eco-Friendly Larvicidal Potential Of Annona Squamosa Ethanolic Leaf Extract Against Anopheles Subpictus

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Abstract

The botanical insecticides are harmless to the environment and living organisms. *Anopheles subpictus* considers crucial research as it searches for innovative solutions to control the disease-carrying vector. Experiments were done in the laboratory to evaluate the larvicidal efficience of ethanolic leaf extract of *Annona squamosa*. The primary metric used to assess the extract's effectiveness was the documented mortality rate of the treated larvae. The median lethal doses (LC50) were (139.117and 113.162) ppm at 24, and 48 hours respectively. These outcomes conclude the effectiveness of *A. subpictus* ethanolic extract as bioinsecticides. Hence, should be incorporated into the integrated pest management strategies as an ecofriendly botanical extract.

Key words: *Annona squamosa*, Natural larvicides, Ethanol extract, *A. subpictus*.

INTRODUCTION

The earliest had installed various control techniques, along with the application of inorganic and organic elements (Jitendra et al. 2009). It was necessary to find alternative biocontrol agents, hence the application of bio-pesticides as natural plant parts/products to withstand the destructive effect of synthetic chemicals. Botanical extracts had become prevalent as a pest control strategy owing to their least persistence, minimum harmfulness to plants and animals, degradability, inexpensive and easily accessible (Senthil and Kalaivani 2005). Plants are a huge store of natural chemical ingredients produced to shield them from pest attacks. Such substances may cause significant physiological disturbances in the life stages of an insect (Rembold 1994). In excess of 30,000 secondary metabolites produced by plants have been identified (Wink 1988). Certain commercial pesticides made from plants (Pyrethrum, Ryania, and Rotenone), were significant feeding deterrents, contact toxins, and active growth inhibitors of some insects (Akhtar et al. 2008). Although pyrethrum is the best-known natural insecticide, it is not appropriate for outstanding surface handlings due to its instability towards the sunlight or strong artificial light therefore, additional synergists are required to increase its toxic effect such as piperonyl butoxide. Akhtar et al. (2008, 2012). Therefore, it is necessary to examine many botanical sources that have high biocidal efficiency to overcome the problems of commonly used plant extracts. Dracena arborea is an abundantly spread type of the plant family; Asparagaceae (Nwaehujor 2013; Okonkwo 2014). This family is branded by fibrous and frequently tough leaves and tightly crowded leaves. It is typically a tropical plant used as an edging plant for defining

boundaries on land because of its strength of renaissance when a small stem is implanted and abundant all year round. It is a woody persistent a tree from the lily family known as dragon tree, a beautiful ornamental plant that grows in semi-dry environments is the. It is indigenous to South Asia and Africa. The moniker "dragon tree" refers to the fact that some species' secretions resemble dragon blood and that a damaged branch usually results in the growth of two new ones. (Udo 2013). The plant is also applied as reptile repellents, ornamental plants for boundaries, as well as agricultural (Burkii 1985). Additionally, it has been found to have therapeutic value, as well as claims that the plant contains components that are anti-fungal and anti-parasitic (Okunji et al. 1996). It has been discovered that several dracaena species are insecticidal. In his research, Udo (2013) attested to the effectiveness of powdered leaves, bark, and roots in eradicating two types of stored product pests that affect maize and beans (Callosobruchus maculatus). (Sitophilus zeamais) and stated that aqueous and ethyl acetate fractions of leaf extract proved its insecticidal activity and existed protection to stored grains. Prosper et al. (2016) recorded its larvicidal activity against Aedes albopictu. Ukoroije et al. (2019) confirmed its biocidal qualities when used as an extract or powder, and it was highly effective in cockroach control. Alkaloids, tannins, saponins, flavonoids, terpenoids, glycosides, and phenols were among the chemical groups that were found through phytochemical screening in varied concentrations. The bioactivity of these chemical groups is linked to resistance to the insect pest. (Udo 2013; Ukoroije et al. 2019). Numerous studies have been directed to identify new bioinsecticides, seeking active substitutes to combat vector mosquitoes. This study aims to find natural ingredients to make formulations that can be incorporated into the integrated pest management strategies as an alternative to synthetic chemical insecticides and to determine its pathological consequence on the target insect.

MATERIALS AND METHODS

Insect rearing:

A. subpictus was raised in a lab setting at 25–30°C, 80–90% relative humidity, and an 11–13-hour light–dark cycle. Until they hatched, eggs were kept in plastic cups filled with clean, dechlorinated water.

Preparation of the plant Powder:

Fresh plant leaves were cut apart, cleaned under running water to eliminate debris, and then dried in the sun for seven days until firm. The leaves were crushed by hand to create a finely separated powder, which was then dried for eight hours at 600°C in a hot air oven. As stated by Udo (2008a).

Extraction procedure of the prepared powder:

The plant material was fully soaked in 90% ethanol and extracted at ambient temperature. The extract was filtered and concentrated in vacuo using a vacuum rotary evaporator. at 40oC for 8 h (Udo et al. 2004). The concentration and percentage yield of the extract was determined. The concentrated *A. squamosa* extract then was stored in a refrigerator at 4oC until further use and diluted using 70% ethanol for the application.

Bioassay of Larvicidal activity:

The larvicidal potency towards the 3 rd larval instar of *Anopheles subpictus* were evaluated using the immersion procedure (WHO 2005). The extract was used in four different concentrations (75, 250, 300, and 500 ppm). Groups of fifteen early 3 rd instar larvae were moving by plastic droppers into test cups, each filled with 10 ml of water under laboratory conditions. For each concentration, three replicates were performed.

RESULTS

The different doses of *Annona squamosa* ethanolic leaf extract against newly moulted 3rd instar *A. subpictus* larvae was assessed and is shown in Figure 1 and 2. Depending on the extract concentrations and exposure duration, the toxicity values vary. As concentrations and exposure times increased, so did the percentage of dead larvae.

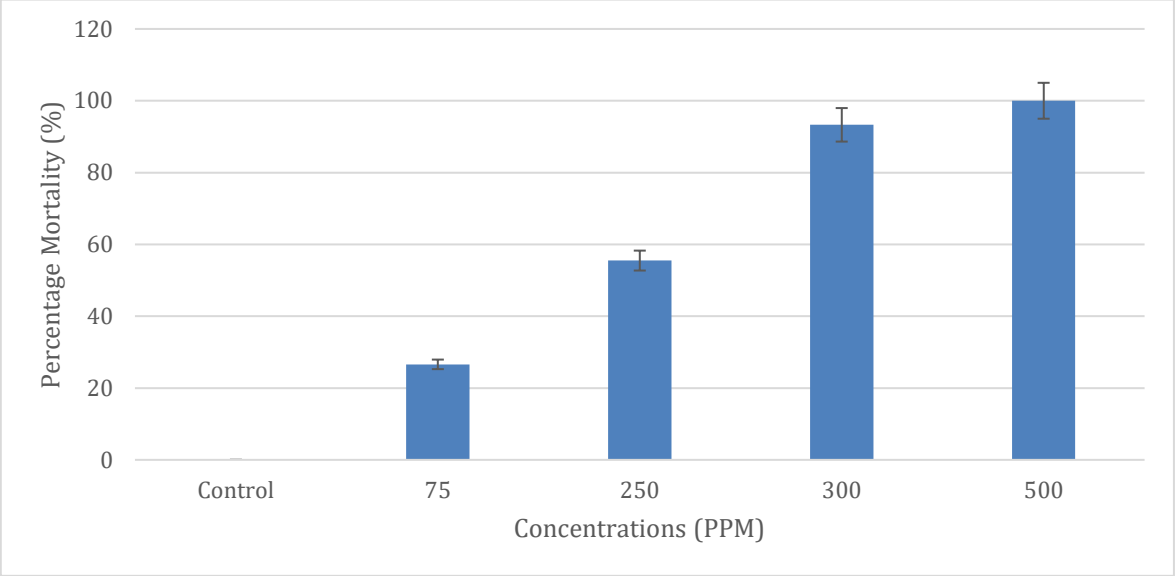

Figure 1: Susceptibility of 3rd instars larvae *A. subpictus* to ethanolic based leaf extract of *Annona squamosa* at different time intervals (24 hrs)

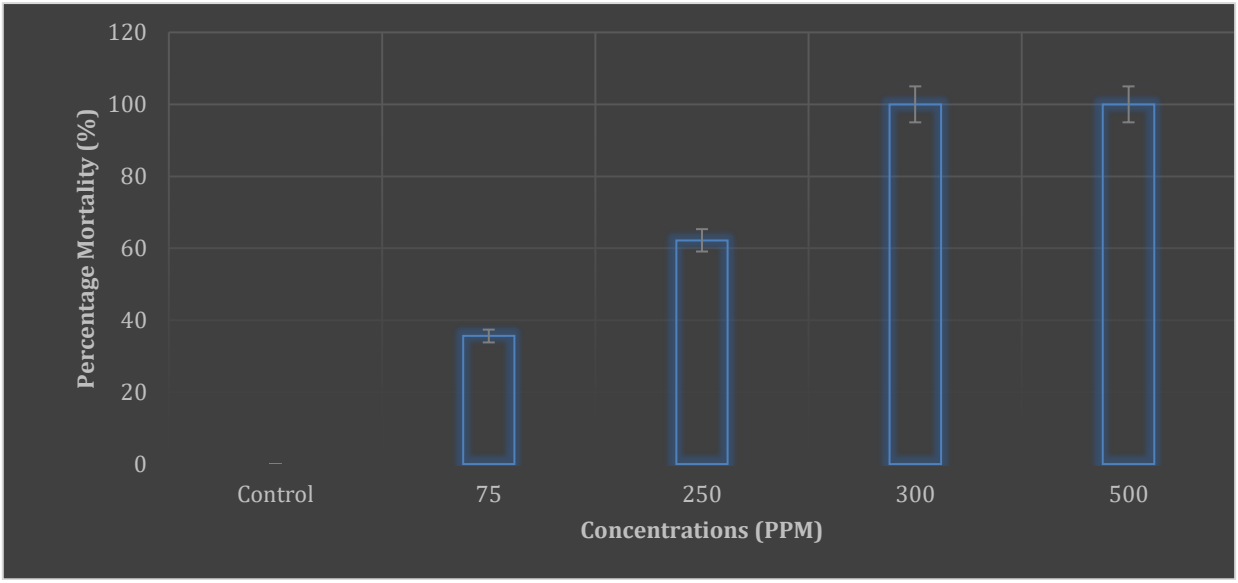

Figure 2: Susceptibility of 3rd instars larvae *A. subpictus* to ethanolic based leaf extract of *Annona squamosa* at different time intervals (48 hrs)

DISCUSSION

The most valuable pest control technique that withstands the environmental hazards induced by synthetic chemicals, and environmentally friendly. Therefore, using natural plants components as biopesticides has been evaluated as the best means of control, has the lowest toxicity to organisms other than the target, is economical, readily available and degradable, and is therefore increasingly being applied (Senthil and Kalaivani 2005). Phyto-extracts are developing as effective pest control agents, that are simple to administrate, low-cost, and risk-free characters. Simple, botanical extracts used as insecticides in numerous nations for ages (Crobsy et al. 1971). Often, complex combinations of active substances make up extracts (Berenbaum 1985). In order to employ indigenous plants as a natural product for mosquito control measures, they must first be tested for mosquito larvicidal activity (Bowers et al. 1995). Phytochemical screening by Ukoroije et al. (2019) listing the existence of chemical groups such as flavonoids, tannins, saponins, terpenoids, alkaloids and glycosides in plants, he explained that these chemical groups have a biological activity against insect's pest. Udo (2013) noted that tannins, saponins, anthraquinones, flavonoids, terpenes, alkaloids, and saponins were all examined using thin layer chromatography to determine the chemical composition of an ethanolic extract of *A. squamosa*. Okunji et al. (1996); Momeni et al. (2005) provoked that the occurrence of harmful secondary metabolites in *A. squamosa* may be the cause of the substantial insect

death. Some secondary metabolites are known to have insecticidal and antifeedant properties, as had been observed by Nawrot et al. (1988); Hassanali and Lwande (1989) against Tribolium castaneum and some lepidopteran pests. In conclusion, some of the *A. squamosa* leaf extract fractions exhibited toxicity, had repulsive effects, and decreased the number of offspring of various insect species. However, it was discovered that the susceptibility to the same plant extract might vary substantially amongst even closely related species (Akhtar et al. 2012). Ethanol is the solvent that has proven to be the least toxic compared to acetate and acetone. In addition to its qualities that repel insects, it is soluble and therefore has a non-polar (hydrophobic) and a polar (hydrophilic) end. The presence of oxygen forms hydrogen bonds and high electronegativity, maximally extracting the active elements present in the test plant sample (Khalequzzaman and Sultana 2006). In the current study evaluation of the toxicity induced through application of the leaf powder ethanolic extract of D. arborea at different concentrations was estimated through recording LC50 and LC59 against the Cx pipiens 3 rd larval instar. As revealed from the results, 50% mortality was obtained at concentrations (113.162) ppm at 48 hrs. Such outcomes indicate its larvicidal activity which is directly proportionated with the concentration and the time of exposure to the extract. The present investigation agrees with Udo et al. (2011) who reported that D. arborea possess larviciding and ovicidal properties in controlling both (C. maculatus) and (S. zeamais), the extract fractions were efficient in decreasing the progeny in each of them. Also, Prosper et al. (2016) recorded its larvicidal activity against Aedes albopictu. Ukoroije et al. (2019) revealed its insecticidal properties in case applied as extracts or in powdered form against Periplaneta americana adults. Alterations in the structure of the integument were induced as a result of the application of the tested botanical extract illustrated as cuticle separation from the hypodermis, hypodermal tissue breakdown, and basement membrane obliteration.Similar remarks were noticed by Younes et al. (1999) and Khalaf et al. (2009). These comprise valuations of minimum environmental and human dangers, shelf-life, their storage resources, the related charges of utilization, and local vector susceptibility (Samuel et al. 2016). Results verified that D. arborea leaf extract had potential larvicidal activity and histopathological effect on *A. subpictus* larvae. In terms of these criteria, botanical products need further investigations for their effectiveness and residual action under field conditions. So, Progressive research is ongoing to detect the active constituents and the mode of action of our tested extract as an environmental bioinsecticide.

CONCLUSION

The current research proved that *Annona squamosa* ethanolic leaf extract contains insecticidal qualities. Its application against *A. subpictus* have added to the enormous source of botanicals utilized as mosquitocidal. The plant-based pesticides are widely distributed and available round the year. Therefore, the use of *A. squamosa* could be an important supplement to synthetic pesticides and possibly will be used as part of integrated pest management plans.

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