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Molluscicidal Effects of *Talinum triangulare on Bulinus truncatus.*

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Abstract
The molluscicidal effects of *Talinum triangulare* from two locations in Nigeria on *Bulinus truncatus* were studied in the laboratory. Snails were exposed for 96h to different concentrations of ethanolic extract of the plant root from Nsukka in Enugu State and Erei in Cross River State, Nigeria. Those in dechlorinated water served as control. On coming in contact with the test medium, the snails reacted by speedily crawling out of the containers. Exposure of snails to *Talinum triangulare* concentrations of less than 300ppm showed only ovicidal activity while varying numbers of those exposed to 300ppm died as the exposure time increased. The control group recorded no effects. Snail recovery was only observed in concentrations less than 300ppm. The LC_{50} of the plant root extract from Nsukka in Enugu State and Erei in Cross River State decreased (ranging from 505-251ppm) as exposure time increased. The molluscicidal activities (LC_{50}) of the ethanolic plant root extracts from the two locations were not significantly different ($p > 0.05$). The ethanolic plant root extracts of *T. triangulare* may be a potential molluscicide in schistosomiasis control.

Key words: Molluscicide, *Talinium triangulare, Bulinus truncatus,* Schistosomiasis.

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Introduction

The phylum Mollusca is the second largest in the animal kingdom after the arthropods with the population of living species being 120,000. Molluscs are highly successful in terms of ecology and adaptation, with representatives in virtually all habitats (Godan, 1983; Ghiselin, 2008).

The larvae of parasitic trematodes pass part of their life in fresh water. Many aquatic snails act as vectors for the larvae of these trematodes and thereby transmit a number of diseases (Singh *et al.*, 2009). Schistosomiasis is a parasitic disease caused by a trematode, *Schistosoma* (a blood fluke). The disease affects 220milion people in endemic developing countries across South America, Africa and the Far East (WHO, 1993), and is frequently referred to as the second most important parasitic disease after malaria among other infectious diseases of the tropical and subtropical countries, and the third most prevalent parasitic disease in the world in terms of overall morbidity, burden, socio-economic and public health importance and human impact (El-Sherbini *et al.,* 2009). Man, the definitive host, acquires infection by contact with freshwater infested with schistosome cercariae, which actively penetrate his intact skin and subsequently develop to the adult worms. These cercariae are released into water by infected snails, in which the parasite undergoes asexual larval multiplication. Snails become infected by miracidia released from schistosome eggs which reach freshwater with human excrement (Adedotun and Odaibo, 2009).

With probably 500,000 deaths per year out of over 220 million people estimated to be infected in endemic countries worldwide, the World Health Organization recommends an integrated control of schistosomiasis. Thus, an appropriately targeted snail control using molluscicide(s) is an important preventive strategy which should be combined with carefully managed and directed chemotherapy, ecological and biological control methods, as well as socio-economic improvements and advances in health education with community participation. Snail control in schistosomiasis is based on the rational assumption that elimination or reduction of snail population density below a certain critical threshold, would reduce transmission to a level at which the rate of new human

infections, as measured by disease incidence, is significantly reduced or stopped altogether (Adedotun and Odaibo, 2009).

The high costs of synthetic molluscicides such as niclosamide, their toxicities to non-target aquatic biota and even man as well as the complex organization required in their application, are a major setback to their continued use, especially in schistosomiasis control programmes. There is thus, the need for cheaper, environmentally friendly, biodegradable and readily available natural molluscicides from plants (Adedotun and Odaibo, 2009; Singh *et al.,* 2010). Plants are the richest source of renewable bioactive organic chemicals. The total number of plant chemicals may exceed 400,000; of these, 10,000 are secondary metabolites whose major role in the plants is reportedly defensive. Plants cause behavioural and physiological effects on pest and vectors of diseases because they possess defensive chemicals of various categories, e.g. terpenoids, alkaloids, glycosides, phenols, tannins, etc. (Singh *et al.,* 2010).

Talinum triangulare or waterleaf, as it is commonly called, belongs to the plant family *Portulaceae*. It is a short-lived perennial herb, growing to 30-60cm in height. The leaf is greenish in colour with succulent stem, alternate leaf arrangement with a tuber-like root. In Nigeria, its wide acceptance across various ethnic groups has earned it several local names, such as 'gare' in Yoruba, 'gbolodi' in Igbo, 'mmon-mmong ikong' in Efik/Ibibio and 'Ebodondon' in Esanland, Edo State (Udoh and Akpan, 2007). It was reported by Okoli *et al.* (2007) in their survey of twenty-one communities (1106 individuals) in Esanland, an ethnic group located in Edo State, Nigeria, that the root of *T. triangulare* taken orally is used by the people to treat schistosomiasis. The present study is a preliminary investigation of the molluscicidal potency of this herb.

Materials and Methods

Collection of snails and plant material: Snails used in the study were collected from Nigercem in Nkalagu, Ebonyi State, Nigeria. Snails were reared in plastic containers containing 3L of dechlorinated water at room temperature and fed fresh lettuce for 14 days. The investigations were carried out according to the methodology described by the World Health Organization for molluscicide screening.

The root tubers of *Talinum triangulare* were collected from Erei in Cross River State and Nsukka in Enugu State, both in Nigeria. The plant roots from the two locations were dried under shade for three weeks, thereafter they were pulverized using a blending machine.

Extraction using organic solvent: Forty (40) grams of the plant root powder from each location were soaked separately in 200mls of ethanol and allowed to stand for 24h at ambient temperature. The ethanolic extract from Nsukka (ETN) and Erei (ETE) were obtained by filtering the soaked materials through Whatman No.1 filter paper and allowed to evaporate to dryness. From the crude extracts of ETN and ETE, stock solutions of 1000mg/ml were prepared using dechlorinated water and different concentrations (25, 50, 100, 125, 200 and 300ppm) were prepared from stock solution. A control test medium was also set up using dechlorinated water.

Molluscicide screening: Molluscicide evaluation of plant extracts were performed according to World Health Organization guidelines for molluscicide screening (WHO, 1961). Ten snails were challenged with 150mls of the different concentrations of ETN and ETE. Each concentration was replicated two times. Snails were exposed for 96h and mortality was recorded after every 24h during the exposure period. Dead snails were removed from test containers to prevent contamination of solution. At the end of 96h, snails were washed and placed in dechlorinated water for a recovery period of 24h.Contraction of body into shell and no response to needle probe were taken as evidence of snail death.

Data analysis: Lethal concentrations (LC_{50}) of the plant root extracts from the two locations were determined using probit analysis (SPSS Version 16.0). Comparing of mean LC_{50} values of ETN and ETE was done using T-test.

Results and Discussion

Snails on coming in contact with the different test concentrations of ETN and ETE speedily crawled out of the test containers. This escape behaviour of snails is in accordance with the work of Jurberg *et al.* (1985), where the authors attributed this movement to the toxic properties of natural and synthetic molluscicides. This behaviour shows an attempt by the snails to escape from an environment whose conditions threaten their survival. Interestingly, it was observed that after few

minutes, snails in 300ppm of ETN and ETE moved to the bottom of the test containers and remained there. This suggests that the concentration in use is near the lethal or sublethal concentration of the two extracts as reported by Jurberg *et al.* (1985). According to Mello-Silvia *et al.* (2006), this response could be as a result of high level of intoxication impairing their movement. At the end of the first 24h, snails in 300ppm test containers of ETN and ETE retracted into their shell and became motionless. As the exposure period increased, dead snails in 300ppm test containers were observed to have swollen cephalopedal mass which failed to respond to mechanical stimulation by blunt object.

The toxicity of ETN and ETE was concentration dependent. No snail mortality was recorded during the 96h observation period in test concentrations of less than 300ppm. Snail mortality increased as the exposure period increased in test concentrations of 300ppm for both ETN and ETE. At the end of 96h observation period, snails were placed in dechlorinated water to recover. Snails in test concentrations less than 300ppm of ETN and ETE recovered after 24h while none in 300ppm recovered. The result showed positive relationship between exposure period and mortality at 300ppm. The time dependent effect of extracts may be due to the uptake of the active moiety which progressively increased the amount of active component in snail body with increase in exposure period or it is possible that the active component(s) could change into more toxic forms in the aquarium water or in the snail body by the action of different enzymes (Singh *et al.,* 2010). Although concentrations less than 300ppm caused no snail death, it was remarkably observed that all the concentrations of ETN and ETE used had ovicidal activity as the egg masses on the shells of the snails were observed to have disintegrated with time before the end of the experiment. No death or ovicidal activity was observed in control group.

The LC_{50} of ETN and ETE is shown in Fig 1. The LC_{50} of ETN decreased as the exposure period increased while that of ETE decreased at 48h, remained steady between 48 and 72h and dropped at 96h. From the figures, it could be deduced that the concentration of ethanolic extract of *T. triangulare* plant roots that will kill 50% of *Bulinus truncatus* 24h after contact would be between 400-600ppm. Mott (1987), however, reported that for a plant to be considered a molluscicide, it should be registered in concentrations up to 100 mg/l and be able to kill 90% of the snails 24 h after contact. On the other hand, Santos *et al.* (2003) using the method of Farnsworth *et al.* (1987) reported the molluscicidal activity of ethanolic root extract of *Physalis angulata* as being weakly active at a concentration of 500mg/l. This therefore implies that the ethanolic root extract of the test plant has a weak molluscicidal activity. Comparing the mean LC_{50} values of ETN (333.28 ± 66.13) and ETE (422.61 ± 77.53) showed that they do not differ significantly($p>0.05$). (According to Azare *et al.* (2007), one of the problems encountered in the use of plant extracts as molluscicides is the choice of solvent for extracting plant material. This suggests that the plant root if extracted with other solvent could have high toxic molluscicidal activity.

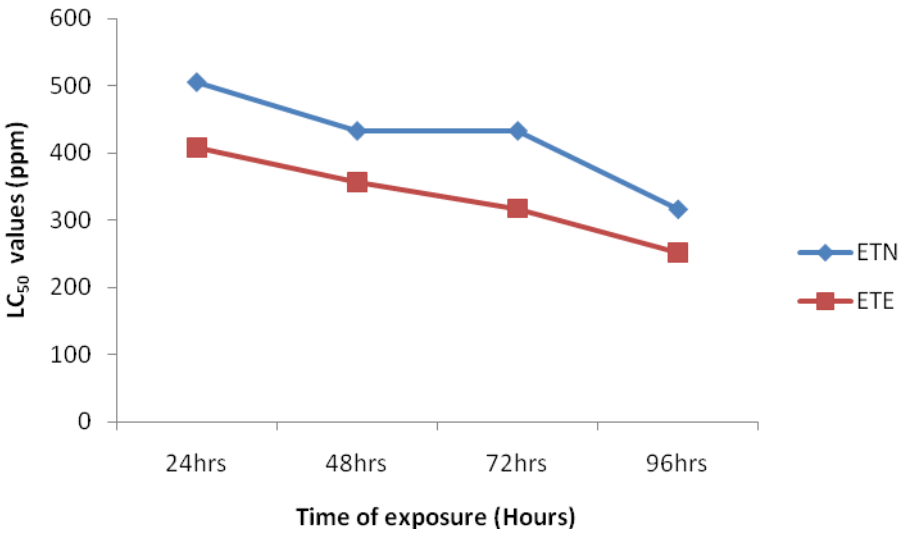

Figure 1: A plot of LC_{50} of ETN and ETE against exposure time.

Conclusion

Plants like *Ambrosia maritima, Jatropha spp, Zingiber officinale, Solanum nigrum, Tetrapleura tetrapleura* and a host of others that have been reported to have anti-schistosomal potency have also been shown to have weak, moderate or active molluscicidal activity on the intermediate host of schistosomiasis. Since report has it that *Talinum triangulare* is used in treating schistosomiasis in Esanland, Benin city, Nigeria, the bioassay of the molluscicidal potency of this plant material using other solvent is strongly recommended. Even though the ethanolic extract of the plant root was observed have a weak molluscicidal effect on *B. truncatus,* the plant material can be evaluated on other biota in the freshwater ecosystem where *B. truncatus* is found and if not harmful to non target animals, be integrated as one of the plant molluscicides used in schistosomiasis control.

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