



Original Article

MOLLUSCIDAL POTENTIALS OF *Opuntia ficus-indica* (CACTUS PLANT) ON *Biomphalaria pfeifferi* AND *Bulinus globosus* (MOLLUSCA: PLANORHYNCHIDAE)

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ABSTRACT

The use plant molluscicides have received considerable attention in the search for cheaper alternatives to chemotherapy and synthetic molluscicides in schistosomiasis control. Molluscicidal activities of *Opuntia ficus indica* (Cactus) extract was tested on *Biomphalaria pfeifferi* and *Bulinus globosus*, the intermediate hosts of *Schistosoma mansoni* and *Schistosoma haematobium*, under standard laboratory conditions. The molluscicidal activity was assessed by determining the efficacy of various graded concentrations (0, 8, 16, 32, 64 and 128 mg/L) of the aqueous plant extracts on the two snail species. Ten (10) adults of uniform size were immersed in 6×3 (replicated thrice) trough. From the result, the highest molluscicidal activities were recorded at 128 mg/L on both snail species. *B. pfeifferi* had the highest mean mortality 6.33 among both species at 24 hours exposure period. At 72 hours the mean mortality of both species were almost the same, however, the highest potency was recorded for *B. pfeifferi*. The LC_{50} and LC_{90} of the *O. ficus* recorded on *B. globosus* were 12.37mg/L and 6.91mg/L respectively while LC_{50} and LC_{90} recorded on *B. pfeifferi* were 5.28mg/L and 1.95mg/L respectively. The result obtained showed that *Opuntia ficus-indica* is a promising plant molluscicide candidate.

Keywords: *Opuntia ficus-indica*, Potency, Molluscicide, Extracts.

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INTRODUCTION

Schistosomiasis is one of the most important public health problems after malaria (Muhammed *et al.*, 2009). It's a

widespread disease infecting over 240 million people (Fenwick *et al.*, 2006). It is found in tropical countries in Africa, Caribbean, Middle East, South America and South East Asia. However an

estimated 85% of the world's cases of schistosomiasis are in Africa, where prevalent rates can exceed 50% in local populations (Hamed, 2006). Out of the infected, over 120 million are symptomatic while over 20 million have severe disease (Steinmann *et al.*, 2006). A further 779 million are at risk of infection in tropical and subtropical areas of the world (Liu *et al.*, 1997). Fresh water snails plays vital role in the transmission of schistosomiasis. Other factors include: lack of sufficiently trained health workers, inadequate safe water Supply, high cost of chemicals for snail control and therapeutic drugs (Amita and Singh, 2001). Schistosomes, the causative agents of the disease have an indirect life cycle requiring aquatic snails, as intermediate host, for their transmission to man. Thus reduction of the parasite transmitting snails plays an important role among the strategies to control the disease (John 2004). Schistosomiasis can be controlled by destroying the carrier snail and thus breaking the life cycles of the parasite (WHO 2004). Snails of the genus *Biomphalaria* are the intermediate host for *Schistosoma mansoni* with the group *pfeifferi* being the most common in Nigeria.

Molluscicides are not frequently used as a major control mechanism due to cost implications and toxicity effects on non-target organisms such as fish (WHO 2004; Adedeji and Okocha, 2012). Therefore there is need for search for a cheap, non-toxic and safer molluscicide (Molgaard *et al.*, 1999; Mossoud and Habib, 2003). Consideration of cost and environmental effect of most molluscicides in current use for the control of schistosomiasis has generated the search for cheaper and less polluting molluscicides from natural source, especially those of plant origin

(Osuna-Martinez, 2014) with the hope that plants showing molluscicidal properties could be used on self-help basis to control diseases in rural areas.

Cactus (*Opuntia ficus-indica*) commonly known as prickly pear belongs to the family Cactaceae. *Opuntia ficus indica* produces sweet, nutritionally rich edible fruits; its tender cladodes are used as fresh green vegetable and salad (WHO 2004). The plant has been used in traditional folk medicine because of its role in treating a number of diseases and conditions, including diabetes, hypertension, hypercholesterolemic, rheumatic pain, gastric mucosa diseases and asthma, in many countries over the world (Reyes-Aguero 2005). Previous researchers have focused investigations for studying genus *Opuntia* in order to discover the properties of plant that could form the basis of their use in the prevention and cure of chronic diseases. Therefore clinical pharmacologic interest in the efficacy and safety of the phytochemicals present in genus *Opuntia* has grown during recent years due to the realization that many people self-medicate using this plant. The aim of this study is to evaluate the molluscicides potential of *Opuntia ficus indica* leaf extract on *Biomphalaria pfeifferi* and *Bulinus globosus* snails.

MATERIALS AND METHODS

Animal material

This study was conducted on Adult stages of *Biomphalaria pfeifferi* and *Bulinus globosus* snails of different diameter. In the laboratory, snails were identified to species level using identification keys by Otarigbo *et al.* (2013). Adults snails were maintained in separate plastic containers with capacity of about 1000ml of

dechlorinated water with stocking density of 10 snail/L at room temperature (25-30°C). *Green Lactuca sativa* was immersed in boiling water for about one minute and then cool with cold water and were used to feed the snails in reasonable quantities at 2 days interval and water was changed twice a week, according to need. Dead snails were removed when observed.

Plant extraction

Opuntia ficus indica whole plants were collected within the premises of Federal University of Technology Minna. They were taken to the laboratory in a wet sack (to avoid direct exposure to sunlight which may lead to dehydration (Adetunji and Salawu, 2010)). The plant was rinsed with distilled water to remove dust, sand, unwanted material and the healthy leaf was selected. Identification of the plant was confirmed by specialist in plant identification, in the department of Biological Sciences, Federal University of Technology, Minna, Niger state. The authentic plants were preserved in the herbarium section of Biological Science Laboratory. Three hundred (300) grams of the plant was pounded using mortar and pestle. The liquid extracts were squeezed using sieving net into a beaker. The extracts were concentrated under reduced pressure at 40atm using rotary evaporator.

Bioassay and Molluscicidal Potency Test of Adult Snail

The molluscicidal potency tests were carried out according to the standard method described by Otariigbo *et al.* (2013) and Adetunji and Salawu (2010). One gram (1g) of each crude extracts was dissolved in 10ml of distilled water and was later transferred into 490 ml of distilled water to prepare the stock solution (500ml). The different volumes

of 0.0 (control), 8, 16, 32, 64 and 128mL from the stock solution of the aqueous extract of the plant were added to an equal volume (500 ml) of dechlorinated water in plastic troughs (10 cm depth × 17 cm in diameter), to have working solutions. Then the concentration of each solution was calculated in milligram per Litre (mg/L): 0.0, 16, 32, 64, 128, and 256 mg/L. Ten (10) adults of uniform size were immersed in 6×3 replicates. In each set up, the snails were prevented from crawling out of the troughs by means of a fine mesh white cloth used for cover and tied to trough by rubber band. The snails were not fed during the course of the experiment; it had been observed that healthy snails live up to five days or more without food (Swaleh and Salawu, 2010), provided other environmental conditions are constant. After 24 hours exposure to the different plant extract concentrations, the snails were transferred to fresh dechlorinated water and maintained there for another 24 hrs. Molluscicidal test with the plant extract doses were separately repeated twice. Death of the snails was determined and confirmed by the lack of reaction to irritation of the foot with a blunt wooden probe to elicit typical withdrawal movements, mortality counts were recorded.

Data Analysis

Descriptive statistical analysis was performed using mean ± standard error of mean. The results relating to molluscicidal activities of the plant extract and tested concentrations were evaluated using Analysis of Variance and Duncan Multiple Range Test, with a significance level of $\alpha = 0.05$. The median and Upper lethal concentrations were determined by regression analysis. All the analysis was carried out using Statistical Packages for Social Sciences (SPSS) 20th version and

Microsoft excel2010.

RESULTS

The Molluscicidal potency of *O. ficus-indica* on *B. globosus* snail species is presented in Table 1. The plant extract showed a significant mortality as the time of exposure moved from 24 hrs to 72 hrs. At 24 hrs exposure period, the mortality recorded was 8 mg/L (1.00 ± 0.00). There were no significant difference in the mortality recorded for 16 mg/L, 32mg/L and 64mg/L at 24 and 48 hrs exposure period. The mortality recorded for all concentration was significantly higher ($P < 0.05$) than that of control. This was same after 48 hrs exposure period. At 48 hours control (0.67 ± 0.07), 8mg/L (2.33 ± 0.33) and 32mg/L (3.33 ± 0.88) recorded no significance different in snail mortality ($P > 0.05$). At 72 hours exposure period, the highest effect on snail was recorded at 128 mg/L (8.33 ± 0.33), however, this was not significantly difference from snail mortality recorded for 64 mg/L (7.67 ± 0.33).

The results on Table 2 showed the Mean Mortality of *B. pfeifferi* Adult E exposed to *Opuntia ficu-indica* extract over a period of 72 hours. At 24 hours, control (0.33 ± 0.03) snail mortality was not significantly different from 8 mg/L (1.33 ± 0.67) at $p > 0.05$, 128 mg/L (6.33 ± 0.33) had the highest mollusc mean mortality and was significantly different from all other concentration at $p < 0.05$. Also at 48 hours 128 mg/L (8.33 ± 0.33) recorded the highest snail mortality and was significantly different from all other mortality recorded at 48 hrs exposure period.

The medial (LC_{50}) and upper (LC_{90}) lethal

concentration of the effect of *O. ficus* extract was presented in Table 3. The LC_{50} and LC_{90} of *O. ficus* against *B. globosus* and *B. pfeifferi* were (6.91, 12.37) and (1.95, 5.28) respectively. The R^2 values recorded confirm the dependence of the mortality observed to the increment in the plant extract

Table 1: Mean Mortality of *B. globosus* Adult snail Exposed to *Opuntia ficus indica* Extract

Concentration (mg/ L)	Hours		
	24	48	72
CONTROL(0)	0.00±0.00 ^a	0.67±0.17 ^a	0.67±0.07 ^a
8	1.00±0.00 ^b	2.33±0.33 ^a	4.67±0.67 ^b
16	1.33±0.33 ^b	3.33±0.88 ^{ab}	3.67±0.88 ^b
32	1.67±0.33 ^b	4.67±0.88 ^b	5.67±0.33 ^b
64	2.67±0.33 ^b	5.67±0.88 ^b	7.67±0.33 ^c
128	4.00±0.58 ^c	6.33±0.33 ^b	8.33±0.33 ^c

Values with the same superscript in the same column are not significantly different at $p>0.05$

Values are presented in mean±standard error of mean of two determinations

Table 2: Mean Mortality of *Biomphalaria pfeifferi* Adult snail Exposed on *Opuntia ficus-indica* Extract

Concentration (mg/ L)	Hours		
	24	48	72
Control(0)	0.33±0.03 ^a	0.67±0.17 ^a	0.67±0.58 ^a
8	1.33±0.67 ^a	2.67±0.88 ^{ab}	4.00±0.58 ^b
16	2.33±0.33 ^b	4.33±0.88 ^b	5.00±1.00 ^b
32	3.00±0.00 ^b	4.33±0.88 ^b	6.00±0.58 ^{bc}
64	3.67±0.33 ^b	5.33±0.88 ^b	7.67±0.88 ^c
128	6.33±0.33 ^c	8.33±0.33 ^c	8.67±0.33 ^c

Values with the same superscript in the same column are not significantly different at $p>0.05$

Values are presented in mean±standard error of mean of two determinations

Table 3: Lethal Concentrations of the *Opuntia ficus-indica* Extract on Adult snails

Snail species	LC ₉₀	LC ₅₀	R ²	Regression Equation
<i>B. globosus</i>	12.37	6.91	0.909	$Y=7.333x-0.666$
<i>B. pfeifferi</i>	5.28	1.95	0.999	$Y=12x+26.66$

DISCUSSION

The environmental and economic problems associated with the use of synthetic molluscicides encouraged the search for substitutes of plant origin. At present, increasing attention is currently given to the study of plant molluscicides in hope that they may prove less toxic, cost-effective, readily available and easily applicable by simple technique. So, the present study is intended to search for ideal molluscicide of plant origin on the basis of dosage response for combating adults snail intermediate host of Schistosomiasis.

From the established regression lines of the tested extract, the molluscicidal activity was similar to that illustrated by Ebenso (2004) and Iglesias *et al* (2008), against terrestrial land snail; Ederly *et al* (1998), against aquatic snails including *B. alexandrina*, this could be as a result of slow stimulus response to the extract as indicated in their research. Furthermore, the toxicity of the tested snails has been shown to increase as the concentration increases. The observed sensitivity of adult towards higher concentration is consistent with Stednichenko (2010) and Kirichuk *et al* (2010). As reported by several investigators, elevated plant extract concentrations have been associated with an aggravated toxic response for several aquatic species.

The susceptibility of *Biomphalaria* to the plant extract when compared to *Bulinus* is in agreement with the work of Munnert *et al* (2003). As reported by many investigators (Abdel-kader and Sheraf, 2000; Abdel-Hamid, 2003), many invertebrate appear to be more tolerant to some substance. From the present

study, it would appear that extract bioaccumulation could be particular in the longevity of *Bulinus*. Some species, however, had a much greater tendency to accumulate one toxicant than the other (Munnert *et al*, 2003). Such differences are mainly related to the mode of exposure, rate of adsorption, species differences and the physiological and histopathological effects induced by the tested metals (Sheraf, 2000).

In the present study, the molluscicidal potency of the plant extract on the longevity of *B. pfeifferi* was also evaluated. From the established regression lines, the molluscicidal activity based on LC_{50} values falls within the range of other plants that have been Judged as promising molluscicides (Abdel-kader and Sheraf, 2000; Abdel-Hamid, 2003; De *et al*, 2005). The recorded toxic effects on the longevity of the studied snails mainly attributed to several factors including plant specific differences of the extracted active ingredient, types of extracted products, differences in their mode of action, method of penetration and the behavioral characteristics of the studied animal (Mantawy *et al*, 2005). It is now well established that in many plants including the tested plants, the activity is due to the presence of bioactive phytochemicals including saponin contents (Rawi *et al*, 1996), tannins compounds (Bezera *et al*, 2002), triterpenoid and alkaloid components (Singh *et al*, 2010).

CONCLUSION

The study carried out showed that *Opuntia* has molluscicidal potential against *Biomphalaria pfeifferi* and *Bulinus*

globosus snails which suggest the plant extract as an alternative option to synthetic molluscicides. Further studies are required to determine the mode-of-action of the plant extract and the damage they cause to the snail tissues as well as the side effects that may exist to the wildlife that surround them, through semi-field trials.

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