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GEO-SPATIAL DISTRIBUTION OF FRESHWATER SNAIL INTERMEDIATE HOST AND MOLLUSCICIDAL EFFICACY OF *JATROPHA TANJORENSIS* FOR THE CONTROL OF *BULINUS TRUNCATUS* IN MINNA.

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ABSTRACT

This study was carried out in Bosso and Chanchga Local Government Area of Niger State between November, 2021 to March, 2022 to evaluate the geo-spatial Distribution of Freshwater Snail Intermediate Host And Molluscicidal Efficacy of Jatropha tanjorensis ***(Hospital too far)*** *for the Control of Bulinus truncatus in Minna. Each sampling site was visited between 8:0 am and 12:00 noon in order to avoid coming in contact with the cercaria which are usually shed from infected snails during the day when the temperature rises slightly above 28^0 c. Snail collection was been made for 45 minutes at each of the site, using scooping net (diameter: 18 cm) supported by a frame mounted on a 2-metre long handle. out of different species of freshwater snail collected from three different water bodies selected (Bosso Dam, Gulubi river and Tagwai Dam) only two species was recorded to be an intermediate host of schistosomiasis.These species are;Bulinus spp and Biomphalaria spp. Bulinus. Fresh leaves of Jatropha tanjorensis was collected from settlements in Bosso Local Government Area of Niger State, Nigeria.The plant was identify with the help of a senior Botanist at the Department of Plant Biology, Federal University of Technology, with reference to the standard flora.Voucher specimens were deposited at the department for future reference. The knowledge of their spatial distributions can be used to map the extent and risk of the disease in edemic areas. Hence, the need to search for better and ecofriendly alternative molluscicide, as a result of high cost of synthetic molluscicide, their environmental toxicity to non target organism and their lethal effect in the environments, propelled this study to elucidate the molluscicidal efficacy of n-hexane, ethyl acetate, aqueous and methanolic extracts of jatropha tanjorensis. The phytochemical constituents of the plant extract were determined using standard procedure. The molluscicidal bioassay against Bulinus spp using graded concentrations of the extract ranging from 100 to 500 mg/l was performed following standard protocol. Mortality was recorded after 24, 48, and 72 hrs exposure period. The effect of the plants on the snail architecture was determined histopathologically. The lethal concentration was estimated using probit linear regression analysis. The result indicated that the plants possess bioactive secondary metabolite including flavonoids, alkaloids, tannins, phenol and saponins. Molluscicidal activities increases with increase in extract concentrations and exposure period. Amongst the extracts, n-hexane and ethyl acetate showed the highest molluscicidal activities with LC_{50} of 3.125 mg/L and 2.967347mg/L, after 48 and 72hrs exposure period. The histopathological results showed that the extract caused mild atrophy, increase in lacuna space, tissue was interspersed by lacunae, reduced cellular presentation and the calcium cells are drastically reduced which is more pronounced in n-hexane and ethyl acetate extracts. Findings from this research suggest the application of n-hexane and ethyl acetate extracts as an alternative agents in the control of snail intermediate host of schistosomiasis.*

KEYWORDS:

INTRODUCTION

Many freshwater snails are of clinical and veterinary importance, serving as intermediate hosts of different helminthic parasites of humans and animals Abdulhamid *et al.* (2018). Schistosomiasis is a neglected tropical disease that affects more than 250 million people in tropical and subtropical regions of the world, with sub-Saharan African accounting for approximately 90% of worldwide cases (WHO, 2017). Each schistosome species uses a different snails species

as its intermediate host, hence the availability of a suitable snail host determines the endemicity of a particular species of *Schistosoma.* Snails of the genus *Biomphalaria* and *Bulinus* acts on the intermediate host of *Schistosoma mansoni* and *Schistosoma haematobium* respectively. At least four approaches of controlling infection have been tried at the community level, these are Control of Snails, Public health education ,Sanitation and Community Based chemotherapy using niclosamide (Jocelyn, 2001). Currently there is an increase attention for the use of new plant Molluscicides that are highly effective, rapidly biodegradable less expensive and probably easily applicable simple techniques than synthetic Molluscicides. The use of Molluscicide to eradicate the snail vector is considered the method of choice to eliminate Schistosomiasis (WHO,1985) as several strategies have been used to control snail population such as physical (Mc Cullough, 1986),chemical (Musola *et al.,* 2003) and biological method (Kloos *et al.,* 2001; Rashed, 2002). Various freshwater bodies in north-western Angola harbuor potential intermediate snail hosts for urinogenital schistosomiasis, highlighting the need to map the rest of the country to identify areas where transmission can occur and where control efforts should be targeted Fiona *et al*. (2017). . Most of the diseases are prevalent in the tropics and are termed neglected tropical diseases because their economic and medical burdens affect poorer/neglected people and because their effect is often ignored/neglected because it can seldom be linked directly to deaths Abe *et al*. (2018).). In view of the fore going, an attempt has been made in this study to evaluate molluscicidal effect of leaf extracts of *jatropha tanjorensis* (Hospital too far), a herbaceous plant of euphorbiaceae family and commonly called catholic vegetable,Iyana-ipaja,Lapalapa (Iwalewa etal.,2005). *Jatropha tajorensis* has been use as a vegetable and for the treatment of diabetic in Nigeria (Olayiwola et al.,2004).

Statement of problem

Many freshwater snails are of clinical and veterinary importance, serving as intermediate hosts of different helminthic parasites of humans and animals Abdulhamid *et al.* (2018). In poor countries where schistosomiasis is common, biological control of the snails that serve as intermediate hosts for Schistosoma and Fasciola, appears feasible and cost effective. The World Health Organization has tested thousands of synthetic compounds for the control of the snail host. Although effective, the high cost of these synthetic molluscicides and their negative impacts on the environment have so far proved them to be entirely inappropriate With Tiwari,(2012) the growing awareness of environmental pollution, efforts are being made to discover molluscicidal products of plant origin worldwide Otarigho,(2012) Being products of biosynthesis, Organic extracts are biodegradable in nature .

Justification for the Study

At least four approaches of controlling infection have been tried at the community level, these are Control of Snails, Public health education ,Sanitation and Community Based chemotherapy using niclosamide (Jocelyn, 2001).Currently there is an increase attention for the use of new plant Molluscicides that are highly effective, rapidly biodegradable less expensive and probably easily applicable simple techniques than synthetic Molluscicides. Globally, more than 200 million people are infected with schistosomiasis and an estimated 779 million are at risk of infection predominantly in sub-Saharan Africa Steimann *et al*. (2006).furthermore, to leverage the cumulative benefit of synergistically acting phytochemical mixtures and a selective combination of potent molluscicidal compound from various plant extracts using different solvent might be effective against the Biomphalaria and *Bulinus* snail. Therefore, it is importance to investigate the geo-spatial distribution of freshwater snail and molluscicidal efficacy of *Jatropha tanjorensis* against the snail which serve as intermediate host of this parasitic infection, so as to provide detail information to public health officials and pharmaceutical company; in order to designed an effective control measure for this Neglected tropical disease.

Objectives of the study

The objectives of the study are to;

i. Identify and mapped out the distribution of snail intermediate host of schistosomiasis
ii. .Identify the bioactive compounds in the extracts that are responsible for the molluscicidal efficacy.
iii. Determine the molluscicidal activity of the crude, n-hexane,ethyle acetate and aqueous extract of the plant on the *Biomphalaria* snail at different concentration (100,200,300,400 and 500mg/L).
iv. Determines LC50 of the plant extract on the snail.

v. Determine the histopathological effect of the plant extract on the *Biomphalaria* snail.

Methodology

This study was carried out in two local government namely; Bosso and Chanchager L.G.A, Minna, Niger State of Nigeria. The study area (Minna) is geographically located in longitude 90^{O} 40'N and latitude 6^{O} 30'E which lies in the southern Guinea Savannah Zone of Nigeria and has a sub-humid semi and tropical climate with mean annual rainfall of 1200 and 1300mm.The higher amount of yearly rainfall in Minna, which amount to 90% of total annual rainfall, occurs between the month of June and September. The average temperature is 22C with the highest falling to within 40^{O}C and 30^{O}C in February/March and November/December respectively and average of 29°C during the wet season (Osunde and Alkassoun,1998).

Samples Collection:

Each sampling site was visited between 8:0 am and 12:00 noon in order to avoid coming in contact with the cercaria which are usually shed from infected snails during the day when the temperature rises slightly above 28^{0} c. Snail collection was been made for 45 minutes at each of the site, using scooping net (diameter: 18 cm) supported by a frame mounted on a 2-metre long handle. Manual search with visual inspection and hand-picking was also been employed at the various sites. The collected snails were then transported to the Animal Biology Laboratory of the university for identification and onward studies. In the laboratory,the snails collected were identified using snails identification key to properly note the species to work with. Brown and Kristensen as used by Abe *et al.* (2018). Map of the study area with Niger State was use to map out the study area. The location (Latitude, Longitude and Altitude) of snail collection sites was geo-reference with a hand-held Garmin 12XL Global Positioning System (GPS)receiver (Garmin Corporation, USA).

Maintenance of Snails in theLaboratory

Collected snails were placed in beakers half filled with dechlorinated water, the beakers were exposed to daylight for one hour to allow emergence of cercariae,one that cercariae are considered infected, all uninfected are maintained in separate plastic containers, The identify Biomphalaria species were maintained in ten different troughs of about 10-15 liters capacity,with each trough containing at least ten (10) snails to 5 litres of dechlorinated and well aerated water. The snails were fed with fresh *Lactuva sativa* (lettuce) and the aquaria was maintained by changing the water three times in a week or when necessary and left to acclimatize for at least 4 weeks.

Collection and Processing of Leaf of *Jatropha tanjorensis*

Fresh leaves of *Jatropha tanjorensis* was collected from settlements in Bosso Local Government Area of Niger State, Nigeria.The plant was identify with the help of a senior Botanist at the Department of Plant Biology, Federal University of Technology Minna, with reference to the standard flora.Voucher specimens were deposited at the department for future reference. The leaves was rinsed thoroughly first with tap water followed by distilled water to remove all the dust and unwanted visible particles and dried at room temperature between 25-30 ^{0} c for at least 14 days to ensure proper drying and avoid sunlight in other to prevent the ultra-violet rays of the sun from destroying the chemical content of the plant. The dried leaves was then pulverised using a mortar and pestle into fine powdery form and stored until extraction for the experiment.

Preparation of Crude Leaf Extract of *Jatropha tanjorensis*

The extraction of the powdered plant material was carried out as described by O' Neill *et al*.(1985) and Najafi *et al* .(2007).four hundred grams (400g) of the powered plant sample was percolated in 1600ml of methanol and kept in the shade for 48hrs.the filtrate was harvested in a beaker and concentrated using a rotary vacuum evaporator Olayemi *et al.*(2014).the crude extract was then preserved at -4 ^{o}C .the crude extract was subjected to solvent fractionation. Therefore ,each of the plant's crude methanolic extracts was dissolved in 100ml of methanol in a beaker: after which, the mixture was poured into one capacity separating funnel and successfully partitioning using n-hexane and ethyl acetate. This give three fractions for each crude extract namely n- hexane, ethyl acetate and methanol.

Identification and Mapping out of Distribution of Snail Intermediate Host of Schistosomiasis in the Study Area.

The snails was identify using shell morphology according to Brown and Kristensen as use by Abe *et at*. (2018). This was achieved by holding the snail shell with the apex pointing upward. When the aperture opens to the right, it is termed dextral but when the aperture opens to the left it is termed sinitral. More so, the sculptural marking on the snail shell was considered during identification. Other shell components that were considered during identification include number of whorls, shape of the shell, type of apex and shape of the peristome on the aperture. The snail intermediate hosts identification was confirmed at the Department of Life Sciences.out of different species of freshwater snail collected from four different water bodies, only two species was recorded to be an intermediate host of schistosomiasis. These species are; Bulinus and Biomphalaria. The total snail collected from each water body was and identified and the percentage of the vector and non-vector of schistosomiasis was then calculated.

Determination of Molluscicidal Effect of the Plant Extract on the Snail.

Molluscicidal effect of the plant extract on the snail was performed according to WHO guideline (1965) as adopted by Rawia *et al.,* (2010). Groups of 10 uninfected *Bulinus snails* snails was be placed in plastic trays with 1000 mL of different concentrations of individual, aqeous, crude, nhexane, and ethyl acetate extracts test solution separately. Four different concentrations of each test solution of the plant extracts was being tested, each with four replicates of 10 snails. Control experiments were being performed with dechlorinated water alone (negative control). The plastic trays was individually covered with a fine plastic mesh to prevent the snails from crawling out. Snails exposed to different concentrations of the plant extract were left for observation for 24 hours, 48 hours and 72hours. After 48 hours and 72hours, the plant extract suspension was decanted and the snails was rinsed twice with dechlorinated water and transferred to a new container filled with dechlorinated water and observed for another 24 hours, which would serve as the recovery period, following which the mortality rates was determined. The percentage of mortality was calculated against the concentrations used.

Result

Photochemical parameters of the four extract of *Jatropha tanjorensis (Qualitative data)*

Extract/phytoconstituent	Phenols	Flavonoids	Tannins	Saponins	Alkaloids
Crude	++	+	++	++	+
Hexane	+	++	+++	+	+
Ethyl acetate	++	+	++	+	+
Aqueous	++	+	+	+	+

Key: (-) absent, (+) trace present, (++) moderately present,(+++) highly present.

Molluscicidal activities of J.tanjorensis using n-hexane,ethyleacetate,methanol and aqueous as solvent at different concentration and hour of exposure.

Exposure Period	Extract	Control	100mg/l	200mg/l	300mg/l	400mg/l	500mg/l
24HRS	NH	0.00±0.000[a]	0.00±0.000[a]	1.75±0.258[bc]	3.75±0.250[d]	6.500±0.646[d]	7.250±0.479[d]
	METH	0.00±0.000[a]	0.75±0.250[b]	1.50±0.288[b]	2.25±0.250[c]	3.750±0.250[c]	5.500±0.500[c]
	EA	0.00±0.000[a]	0.00±0.000[a]	0.25±0.250[a]	0.00±0.000[a]	1.250±0.250[b]	2.500±0.500[b]
	AQ	0.0±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.800±0.490[a]	1.600±0.980[a]
48HRS	NH	0.00±0.000[a]	1.75±0.250[c]	4.25±0.250[d]	8.25±0.629[e]	8.75±0.250[e]	10.00±0.000[f]
	EA	0.00±0.000[a]	1.25±0.479[ab]	2.25±0.250[c]	5.75±0.250[e]	9.25±0.250[f]	10.00±0.000[f]
	METH	0.00±0.000[a]	0.00±0.000[a]	0.25±0.250[a]	1.00±0.000[a]	2.00±0.000[b]	3.75±0.250[c]
	AQ	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.500±0.500	1.000±1.000[a]
72HRS	METH	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	2.25±0.250[c]	7.75±0.250[e]	10.000±0.000[f]
	AQ	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]	0.00±0.000[a]

Values are expressed as mean ±SEM of three replicates. The values along the same row with different superscript are significantly different (p<0.0

Percentage of the distribution and abundance of vector and non-vector of schistosomiasis

Names of Water Body	Total Number of Snail Collected	Vectors				Non-Vector	%Prevalence
		Bulinus spp	% prevalence	Biomphalaria spp	% Prevalence		
BOSSO DAM	1025	634	62	368	3	23	2
GULUBI RIVE	634	36	6	23	4	575	90
TAGWAI DAM	772	42	5	27	4	703	91

Discussion

Findings from this study indicated that the methanolic, n-hexane, ethyl acetate and aqueous extracts of the Jatropha tanjorensis contains an appreciable amount of bioactive compounds including flavonids, saponins, tannins, phenols and alkaloids. These compounds are synthesized via secondary metabolic activities by plants in order to defend themselves against infections and pathogens (Kumar *et al.,* 2017). The leaf extract also possesses antimicrobial properties and inhibit the growth of *S. aureus* and *E. coli* (Oboh and Masodje,2009).In the current study, it was generally observed that molluscicidal efficacy was dose and time dependent. This was observed when the extract was applied singly, because the higher the extracts concentration and time of exposure, the higher the rate of snail mortality in the treated

groups. This observation could be attributed to the increase in concentration of the bioactive compounds, which increased the toxicity potentials of the extract as well as bioaccumulation of this toxicant in the snail body as time of exposure increases. This finding is in agreement with the previous reports of Fayez *et al*. (2006) who time dependent molluscicidal potency of *Agave filitera*. Similarly Rawi *et al*. (2011) added that plant molluscicidal potency is time and concentration dependent. Amongst the four plant extracts tested, this study also observed *n-hexane* extract to be more potent against the exposed Biomphalaria snails. These could be attributed to the fact that the preliminary screening showed that the plants possess more secondary metabolites than the other three screened. This could be that the phytochemical caused synergistic effect against the snail species compared to other solvent used in the plant extract. The mortality recorded could be attributed to the presence of phytochemical that was identified during the experiments. This group of secondary metabolites may be responsible for the mortality of snails (Alinsub and Bagot, 2019). It has been well established that Tannin-bearing plants possesses molluscicidal potency and molluscs tends to avoid these plant in the ecosystem. (Molgard, 1986; Schaufelberger and Hostettmann, 1983). Saponins have been mentioned severally as active molluscicide (Bezeira *et al*., 2002; Musman *et al*., 2014) with hemolytic properties. In addition presence of saponins caused deformation of a complex reaction with plasma as well as membrane cholesterols causing cell membrane damages (Moses *et al*., 2014). According to Guruswamy *et al*. (2017), cardiac glycosides decreases acetyl cholinesterase (AChE) activities and impaired the hepatopancrease tissues of snails thereby causing fetal inhibition of the digestive enzymes and feeding rate. This is an indication that these bioactive metabolites while acting singly and/or synergistically could inhibit the activities of detoxification system of the snail, hence the observed mortality. The synergistic action observed in this current study can be considered associated with the combined toxicity of the secondary metabolites of the individual extract. Thus the combination of the lethal actions of the active compounds of the tested plant in the current study possibly caused the increase in the snail mortality recorded..Few studies exist using GIS and spatial statistics to define the extent of the distribution of *Bulinus sp* (Malone et al 1997;Opisa et al 2011).Snail distribution map have been used effectively for schistosomiasis control measure in East Africa (Malone et al 1997).The model maps of spatial distribution of infected *B.truncatus* can be used by control programmed to create intervention buffer and identify communities at risk for intervention,thereby reducing the cost of parasitological survey.Measures should be taken through health education and public enlightenment on the danger of wading in potential infective water bodies.The prevalence studies indicated that *B.truncatus* snail may be wide spread with the potential danger of urinary schistosomiasis transmission in water bodies in minna area of niger state.

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