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Antischistosomal Activity of the Aqueous Leaf Extract of *Vernonia amygdalina* on *Schistosoma haematobium* Infection in Rats

Nnaji Nonyerem Joy *, William Istifanus Aliyu and Adamu Samaila Babayo

Department of Biological Sciences, Abubakar Tafawa Balewa University, P.M.B 0248, Bauchi, Nigeria.

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Abstract

Schistosomiasis remains one of the most debilitating Neglected Tropical Diseases (NTDs), with Praziquantel being the only drug of choice. However, dependence on a single drug is threatened by limited access, high cost, and possible resistance. Hence, it is imperative to search for cheaper and locally available alternatives. This study evaluated the antischistosomal activity of the aqueous leaf extract of *Vernonia amygdalina*, a common Nigerian shrub used both as soup and cure for stomach ailments. *Bulinus globosus* snails were challenged with miracidia obtained from *S. haematobium* eggs. Cercariae from infected snails were used to infect 15 albino rats through a modified tail immersion method. The infected rats were divided into five groups of three rats each. Three groups received100mg, 150mg, and 200mg dosesof the extract, a fourth group was treated with Praziquantel and the last group was left untreated. A sixth group remained uninfected, totaling 18 rats. Treatment with *V. amygdalina* significantly reduced worm load in a dose-dependent manner, with the 200 mg group showing the greatest reduction. Gross pathology revealed that the untreated rats exhibited liver mottling and hepatomegaly, which were markedly reduced at 200 mg. Clinical observations showed hair regrowth, absence of haematuria, and normal grooming comparable to the praziquantel-treated group. Lower doses produced partial improvements.Overall, the aqueous leaf extract of *Vernonia amygdalina*demonstrated potent antischistosomal activity, and could serve as a promising alternative therapy.

Keywords: Schistosomiasis; *Vernonia amygdalina*; Praziquantel; Efficacy

1. Introduction

Schistosomiasis also known as snail fever is one of the Neglected Tropical Disease (NTD) affecting over 200 million people worldwide, particularly in sub-Saharan Africa where poverty, poor sanitation, and limited healthcare access promote transmission (WHO, 2023). In Nigeria, *Schistosoma haematobium* remains endemic in many rural communities, where poverty, poor sanitation, and limited access to potable water increase transmission risk Adeyemo *et al.,* (2025). Control of the disease largely depends on chemotherapy, with praziquantel (PZQ) being the only drug of choice. Although PZQ has significantly reduced morbidity, its long-term effectiveness is threatened by issues of high cost, limited availability in remote areas, and the potential emergence of resistant strains Ojurongbe *et al*., (2014). These challenges emphasize the need for search of alternative chemotherapeutic agent that are locally available, safe, affordable and sustainable in endemic settings. One promising approach is the use of medicinal plants, which are traditionally employed in African communities to treat parasitic infections. Plant based remedies are often locally available, culturally accepted, and comparatively affordable, making them attractive candidates for integration into schistosomiasis management strategies Acheampong *et a*l., (2020). Among such plants is *Vernonia amygdalina* (commonly known as bitter leaf), a perennial shrub widely consumed as a vegetable in Nigeria and other parts of Africa. In addition to its nutritional role, *V. amygdalina* has demonstrated various pharmacological activities, including antimalarial, antidiabetic, antioxidant, and antimicrobial properties Adewuyi et al., (2025). However, given the pressing

*Corresponding author: Nonyerem Joy Nnaji

need for affordable therapeutic options and the ethnopharmacological relevance of *V. amygdalina*, We report here the antischistosomal activity of aqueous leaf extract of *V. amygdalina* in infected albino rats.

2. Materials and Methods

2.1. Collection of leaves sample

Fresh leaves of *Vernonia amygdalina* were harvested from farms in Gwallamejie village, Bauchi, Nigeria. The leaves were placed in a polythene bag and were transported to the herbarium section of the Department of Biological Sciences in Abubakar Tafawa Balewa University for identification and authentication.

2.2. Preparation of plant extracts

The aqueous extract of *Vernonia amygdalina* leaves was processed as described and modified by Okpoghono *et al*. (2018). The leaves of *Vernonia amygdalina* were allowed to shade dry at room temperature on the laboratory bench for 7weeks (owing to high humidity during the rainy season) before being pulverized to fine powder using a prestige blender model MJ-60BM01C. Two hundred gram (200mg) of the leaf powder was measured and then soaked in 250ml distilled water in a 500ml round bottom flask. The mixture was left on an electric shaker for 72 hours so as to ensure proper extraction. The extract was filtered using whatman No 5 filter paper and the filtrate was concentrated to a pasty form by allowing it to evaporate at 50C on Julabo TW20 water bath. The resultant aqueous extract was stored in an air-tight sterile container in a refrigerator until required for use.

2.3. Parasite Source and Infection of rats

Schistosoma haematobium eggs were isolated from infected urine samples collected from Primary School children in Firo, a known urinary schistosomiasis endemic village in Ganjuwa Local Government Area of Bauchi state. Parasite free *Bulinus globosus* were infected with miracidia obtained from hatched eggs and resultant cercariae were harvested and use in infecting 15 albino rats through the modified tail immersion method.

2.4. Clinical observation on infected rats

Several clinical signs were observed in the rats post infection. These included loss of fur, haematuria (blood in urine), increased urinary frequency, and behavioral changes that became more pronounced as the infection progressed. While all the 15 infected rats had haematuria, wet perineal region and weight loss, only half of that number showed loss of fur. The other clinical signs were observed in fewer numbers of infected rats ranging from 3-6.

2.5. Perfusion, worm Recovery and Gross pathology

Adult worms were recovered from infected rats using the hepatic perfusion technique as modified by Viana *et al*., (2018). Infected rats were sacrificed 9 weeks post-infection under chloroform anesthesia. To minimize blood clotting during perfusion, aspirin was administered orally before sacrifice. A cannula was inserted into the left ventricle of the heart, and normal saline at room temperature was gently perfused, until the liver appeared pale and the effluent was clear. Worms flushed from the portal and mesenteric veins were rinsed into Petri dishes, and counted under a dissecting microscope. Gross pathological changes were assessed in the liver and urinary tract. The liver was examined for hepatomegaly, mottling, and discoloration, while the bladder and ureters were inspected for thickening, hemorrhagic lesions. These gross findings were used to assess the extent of pathology and therapeutic efficacy of the aqueous leaf extract of *Vernonia amygdalina* compared with praziquantel and untreated controls.

2.6. Treatment of rats with plant extract

A total of 18 albino rats were divided into six treatment groups (A-F) of three rats each. Groups A, B and C were treated with 100mg, 150mg and 200mg of the extracts respectively as a single dose. In addition, group D was treated with Praziquantel the current drug of choice, while group E was left untreated as the negative control and group F was the uninfected and also untreated group.

3. Results

3.1. Clinical observations in *S. haematobium* infected rats before treatment

Several clinical signs were observed in the rats post infection. These included loss of fur, haematuria (blood in urine), increased urinary frequency, and behavioral changes that became more pronounced as the infection progressed. These

are set out in table 1. While all the 15 infected rats had haematuria, wet perineal region and weight loss, only half of that number showed loss of fur. The other clinical signs were observed in fewer numbers of infected rats ranging from 3-6. The various clinical signs observed during the study are consistent with signs systemic illness reported in schistosomiasis models and agrees with the report of Acheampong et al., (2020) in their work on schistosomicidal and cercaricidal activities of some selected medicinal plants from Ghana. Altogether, these clinical findings provide evidence of morbidity induced by *S. haematobium* infection, and suggested the successful establishment of the experimental model.

Table 1 Clinical observations recorded in *S. haematobium* infected rats before treatment

Clinical Signs	**Descriptions**	**Number of rats involved N=15**
Loss of fur	Dull and rough coat	8
	Patchy hair loss	7
Abnormal behavior	Huddling	5
	Poor feeding	6
	Reduced grooming	4
Reduced activity	Weakness	3
	Prolonged inactivity	5
Haematuria	Passing of blood in urine	15
Wet perineal region	Wetness around perineal region	15
Weight loss	Decrease in weight	15

3.2. Gross pathological effects observed in schistosome infected rats before treatments.

Post-mortem examination of the infected untreated rats revealed distinct gross pathological signs associated with *S. haematobium* infection. The major findings included hepatomegaly, liver mottling, as well as multiple granulomatous lesions in the stomach, ileum, colon, and caecum. These pathological signs are shown in Table 2.

Table 2 Gross pathological effects observed in schistosome infected rats before treatment

Pathological signs	**organ**	**Severity**
Hepatomegaly	Liver	+++
Mottling /discolouration	Liver	+++

Key: + = trace; ++ = Moderate; +++ = severe

3.3. Worm Load

The infected untreated control group recorded an average worm load of 56.1 worms, all of which were alive. This represented the baseline worm burden in the experimental rats before treatment effects were considered.

Table 3 Mean worm load recovered from infected untreated control rats

Worm recovery	**Mean worm count**
Worms recovered alive	56.1
Worms recovered dead	0.0
Total worm load (alive +dead)	56.0

3.4. Clinical impact of different doses of treatment on infected rats during the study

Table 4 shows the clinical impact of the treatment on infected rats. It can be seen that the 200 mg treated group showed the best recovery, with all rats (3/3) exhibiting complete hair regrowth, absence of haematuria, and normal grooming activity, comparable to the praziquantel group. At 150 mg, partial improvements were observed, with one rat showing hair regrowth and reduced haematuria, while the 100 mg group showed only slight changes in grooming activity and there was no hair regrowth. In contrast, the infected untreated group had persistent haematuria, no hair regrowth, and reduced grooming activity, whereas the uninfected group remained normal.

Table 4 Clinical impact of different doses of treatment on schistosome infected rats during the study

Clinical observations	**Treatment Groups**				**Infected untreated +ve control (n=3)**	**Uninfected –ve control (n=3)**
	100 mg (n=3)	**150 mg (n=3)**	**200 mg (n=3)**	**Praziqantel (n=3)**		
Haematuria	3/3	2/3	0/3	0/3	3/3	0/3
Huddling	2/3	2/3	0/3	0/3	3/3	0/3
Grooming activity	1/3	1/3	3/3	3/3	0/3	3/3
Wet perineal region	2/3	1/3	0/3	0/3	3/3	0/3
Weight gain	Minimal	Moderate	Marked	Marked	poor	
Hair regrowth	0/3	1/3	3/3	3/3	0/3	3/3

3.5. Effects of treatment with different doses of V amygdalina on worm recovery

The effect of treatment with the *V. amygdalina* extract on worm burden in infected rats is presented in Table 5. The infected untreatedgroup (+Ve control) had the highest average number of worms recovered alive of 56.1 (100%) while the 200 mg treated group had the lowest number of worms recovered alive being 8.7 (15.5%). The least effective treatment was observed in the 100 mg which had a live worm recovery of 29.6 (52.8%). As compared to treatment with praziquantel which had a 13.2% live worm recovered.

Table 5 Worm recovery after treatment with different doses of V amygdalina on infected Rats

Dosage	**Average Worm Recovery**		**Efficacy (%)**
(mg)	Worms Alive	Worms Dead	
100	29.6	26.5	47.2
150	22.8	33.3	59.4
200	8.7	47.4	84.5
Pz	7.4	48.7	86.8
Infected and untreated	56.1	100	N/A
Uninfected	0	0	N/A

Key: N/A= Not applicable

3.6. Effect of treatment with different doses of *V amygdalina* on Liver mottling

The effect of treatment with varying doses of *V. amygdalina* aqueous leaf extract is shown in table 7. The result show that the infected untreated group had the highest liver mottling score 19.3. In contrast, the 200 mg extract treated group showed a marked improvement with a low score of 5.6 representing a 70.9% reduction in liver mottling compared to the +ve control group. This implied that treatment with 200 mg of the extract significantly reduced liver damage relative to the untreated infected control.

Table 6 Liver mottling scores after treatment with varying doses of *V amygdalina* aqueous leaf extract

Treatments (mg)	Liver mottling scores	Percentage reduction in mottling (%)
100	8.6	55.4
150	8.0	58.5
200	5.6	70.9
Pz	5.0	74.1
Infected and untreated	19.3	N/A
Uninfected	0	N/A

4. Discussion

The various clinical signs observed during the study are consistent with signs of systemic illness reported in schistosomiasis models and agrees with the report of Acheampong et al., (2020) in their work on schistosomicidal and cercaricidal activities of some selected medicinal plants from Ghana. Altogether, these clinical findings provide evidence of morbidity induced by *S. haematobium* infection, and suggested the successful establishment of the experimental model.

The pathological signs observed in this study demonstrate the extensive organ damage caused by *S. haematobium* infection in experimental rats. The liver changes hepatomegaly, mottling, and discoloration reflect severe hepatic involvement due to schistosome egg deposition and inflammatory reactions. This closely agrees with the report of Acheampong *et al.* (2020) who similarly observed hepatic enlargement, discoloration and mottling in rodent models of schistosomiasis.

In this study, a mean worm load of 56.1 worms per rat in the infected untreated control group was observed. This finding demonstrated the successful establishment of *Schistosoma haematobium* infection in the experimental rats and provided the baseline against which the efficacy of *Vernonia amygdalina* extract was evaluated. The number of worm recovered from the experimental infection in this study agrees with the figure obtained in an earlier work in Bauchi by Adamu (2004) who recovered a total of 57 worms from his infected rat model. This probably shows the virulence of the local schistosome strain in Bauchi state. However, the total number of adult schistosomes recorded in this study was much higher than those reported by other workers like Ogboli et al., (2000) and Acheampong et al., (2020) who recovered 2 and 14 adult worms respectively. These differences could be associated with the method they adopted in infecting the rats where they injected cercariae suspension into the rats intraperitoneally or percutaneously. In this study, infection of rats was done through the tail immersion method which is apparently a better method of ensuring that the rats models are properly infected and a sizable number of adult worms could be obtained

Thus, the clinical recovery observed in the 200 mg treatment group of *V. amygdalina* highlighted the potential of the extract in ameliorating schistosomiasis-induced morbidity. The outcome in this study reinforces the therapeutic value of *V. amygdalina* as an indigenous medicinal plant especially in the management of schistosomiasis.

The present finding is also comparable with the report of Akinmoladun *et al*. (2019) who demonstrated that *Azadirachta indica* leaf extract significantly reduced worm burden and egg output in schistosome infected rats. This finding corroborates the report of Abongwa *et al*. (2016), who demonstrated that *V. amygdalina* extracts significantly lowered worm and egg burden in *Schistosoma mansoni* infected mice, with improvements in liver histopathology.

This finding is consistent with that of Olorunnisola *et al*. (2013) who demonstrated that *V. amygdalina* possesses antioxidant and hepatoprotective properties, capable of attenuating liver damage induced by oxidative stress in experimental models, As well as that of Adedapo et al. (2014), who observed restoration of normal histological features in the liver and kidney of infected rodents treated with V. amygdalina. Comparable observations have equally been made with other plant-based antischistosomal candidates. For instance, Adenowo *et al.,* (2015) found that Carica papaya seed extract significantly reduced liver granulomatous reactions in schistosome infected animals. Likewise, Akinmoladun *et a*l., (2019) observed that *Azadirachta indica* leaf extract improved liver histology and reduced fibrosis in schistosome infected rats. These studies strengthen the argument for medicinal plants as potential alternatives or adjuncts to praziquantel in reducing hepatic pathology as well as their general usefulness in schistosomiasis management. Variations in efficacy reported across these studies may be attributed to differences in extraction solvents, plant

chemotypes, and experimental models. The strong effects observed in the present study, particularly at 200 mg, suggests that the bioactive compound in *V. amygdalina* extract are potent against schistosome parasites and associated pathology.

5. Conclusion

This study has revealed that *Vernonia amygdalina* aqueous leaf extract exhibits significant antischistosomal activity in experimentally infected rats. The highest dose (200 mg) was the most effective, producing reductions in worm burden, amelioration of gross pathological changes, and restoration of normal clinical outcomes comparable to praziquantel. These findings support its potential as a locally available and affordable therapy for schistosomiasis.

Recommendations

- Further studies should investigate the mechanisms of action of the extract.
- Toxicological studies should establish the safety profile and optimal therapeutic dosage.
- Histopathological investigations should be conducted to detail tissue-level repair and parasite clearance.
- Clinical trials in endemic communities are needed to evaluate effectiveness in humans.

Compliance with ethical standards

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Disclosure of conflict of interest

We declare that there are no conflict of interest in connection with this paper..

Statement of ethical approval

This study was carried out following the ethical guidelines for the use of laboratory animals as approved by the department of Biological Sciences, Abubakar Tafawa Balewa University, Bauchi. All experimental procedures involving rats were performed in compliance with institutional and International standards for the care and use of laboratory animals.

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