



Original Article



Freshwater Malacological Fauna of the Ruzizi Plain, Democratic Republic of the Congo: Biodiversity and Outbreaks of Schistosomiasis

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Abstract

Background: In the Ruzizi plain, intestinal schistosomiasis, caused by *Schistosoma mansoni* and transmitted by *Biomphalaria pfeifferi*, remains a significant public health challenge. This research offers insights into the diversity of molluscan species in the Ruzizi Congolese plain and the prevalence of schistosomiasis.

Methods: The molluscan samples were gathered using a 0.5 mm mesh dip scoop net, sieves, and plastic containers in 2015. Data on schistosomiasis outbreaks from 2010 to 2014 were sourced from local hospitals, supplemented by cases recorded in 2015.

Results: The study collected 3763 snails, representing 11 gastropod species. The most prevalent were *B. pfeifferi* (30.75%), *Melanoides tuberculata* (25.64%), and *Melanoides* sp. (18.89%). Ponds and agricultural irrigation canals contained a higher mollusk abundance than rivers. Schistosomiasis infestation rates were 24.5, 18.3, and 15.4% in Luvungi, Sange, and Kiliba villages, respectively. Canonical correspondence analysis (CCA) revealed positive correlations between *Bulinus forskalii*, *Biomphalaria* sp., and *Bulinus globosus* with pH, and *Melanoides* sp. with electrical conductivity. In contrast, *Bellamya* sp. showed a negative correlation with dissolved oxygen (DO).

Conclusion: The study confirmed schistosomiasis presence across the Ruzizi Congolese plain, with Luvungi exhibiting notably high prevalence. Just *B. pfeifferi* was identified as shedding *S. mansoni* cercariae in the region.

Keywords: Aquatic snail, Infestation rates, *Biomphalaria pfeifferi*, Physico-chemical parameters, Canonical correspondence analysis

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Introduction

Mollusks are ubiquitous in African freshwater ecosystems and often serve as significant hosts for parasites, playing pivotal roles in both medical and veterinary contexts due to the natural transmission of diseases.¹⁻¹⁰ The Ruzizi plain, situated in the Albertine Rift, boasts a rich malacological biodiversity.⁸⁻¹⁸ Despite this richness, there is a notable scarcity of information regarding the freshwater gastropods of the Ruzizi Congolese plain. A previous study in Kiliba from four years ago examined the intermediate host snails of schistosomiasis,¹² but it had a limited scope, not encompassing the entire Ruzizi plain. Intestinal schistosomiasis, stemmed from *Schistosoma mansoni* and transmitted by *Biomphalaria pfeifferi*,¹³⁻¹⁵ poses a significant public health challenge in the Ruzizi plain, with high incidence rates in regions dominated by

agricultural activities.¹⁴ The true prevalence and extent of schistosomiasis in this region remain elusive due to inadequate large-scale studies and insufficient data on infestation rates and disease outbreaks. Stakeholders combatting bilharzia in this area emphasize the critical need for comprehensive insights into the diversity and ecology of freshwater mollusks. A comprehensive evaluation of all freshwater mollusk vectors, both those responsible and not responsible for schistosomiasis, in the Ruzizi area is essential for effectively managing the disease.

This study aimed to: (1) ascertain the mollusks' diversity and abundance in ponds, irrigation canals and rivers across three villages in the Ruzizi Congolese plain; (2) establish models detailing the relationships between snail species and the physicochemical properties of the



freshwater habitats; and (3) evaluate the prevalence of schistosomiasis from 2010 to 2014 in the Ruzizi Congolese plain.

Material and Methods

Study area

The Ruzizi Congolese plain is situated within the coordinates of 2.5°-3°24' S latitude and 28.5°-29° E longitude. The current research was performed in three specific locations: Luvungi, Sange, and Kiliba (Figure 1). The plain is bordered on the northwest by the towering Kivu mountains, on the east by the Ruzizi River, and on the south by Lake Tanganyika. The prevalent vegetation observed across all sites consists of *Phragmites mauritianus* Kunth, 1830; *Typha*; *Nymphaea*; *Ceratophyllum*; *Cyperus*; *Hyphaene ventricosa* Kirk, 1866; and *Acacia polyacantha* Willd, 1806. The selection of these sampling sites was based on factors such as accessibility and human activities in the area.

Sampling Procedure

In 2015, we conducted sample collection activities in various villages, each consisting of two ponds, two irrigation canals and two rivers. A total of 18 collection points were visited, with each point being visited twice. To gather molluscan samples, we utilized tools such as a dip scoop net with a mesh size of 0.5 mm, sieves, and plastic containers. In addition to the sample collection, we obtained medical statistics on schistosomiasis outbreaks between 2010 and 2014 from hospitals in Luvungi, Sange, and Kiliba. Furthermore, we collected data on schistosomiasis cases recorded in 2015.

To understand the factors influencing species distribution, we measured several physicochemical factors, including water temperature (°C), dissolved oxygen (DO) (mg/L), conductivity (µS/cm), and pH. These measurements were taken using a multiparameter apparatus.

Identification of Freshwater Gastropods

In the laboratory, we employed systematic keys based on recognizable shell morphological characteristics^{2,16-19} to identify all mollusk specimens. Among the snails that were captured, our focus was on *Biomphalaria pfeifferi*, which is known as an intermediate host for *S. mansoni*. To determine if the *B. pfeifferi* snails were hosting trematode cercariae, we utilized the emerging method.²⁰ All living snails were placed in a small plastic cup filled with distilled water and exposed to sunlight or intense artificial light for a minimum of four hours. Subsequently, the cup was regularly examined with the naked eye to identify the presence of *Schistosoma* cercariae. If no cercariae were observed within four hours, the snail was considered negative. The infestation rate was then calculated based on the populations of *B. pfeifferi* tested at each site.

Data Analysis

To compare variables, statistical analysis was conducted using ANOVA and Kruskal-Wallis tests in R software (version 4.3.1). These tests were utilized to assess the differences between the variables under consideration. Furthermore, a canonical correspondence analysis (CCA) was performed to visualize the responses of the snail community to the physicochemical parameters. The CCA helps to identify relationships and patterns between the snail community and the measured parameters. Additionally, Pearson correlation coefficients were calculated to examine the correlation between the species abundance of *B. pfeifferi* and schistosomiasis. This analysis provides insights into the potential relationship between the abundance of *B. pfeifferi* and the occurrence of schistosomiasis.

Results and Discussion

Malacological Fauna in the Ruzizi Congolese Plain

A total of 3763 freshwater snails were gathered, comprising 11 species across ponds, irrigation canals, and rivers.

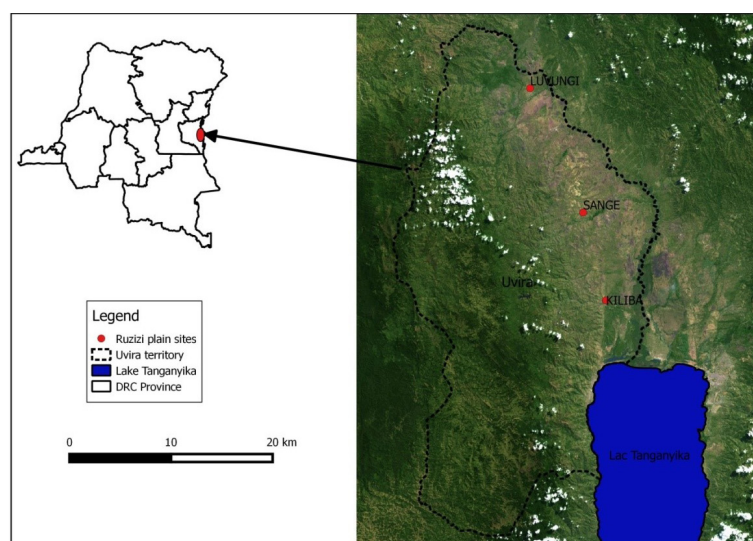


Figure 1. Map Indicating the Sites Sampled in the Ruzizi Congolese Plain

Detailed in Table 1, ponds harbored 9 species, irrigation canals 8, and rivers 5. Ponds exhibited the highest snail count, constituting 46.68% of the total, followed by irrigation canals at 41.59%. Rivers had the lowest count at 11.73%. *B. pfeifferi* emerged as the most prevalent species, making up 30.75% of the population, succeeded by *Melanoides tuberculata* at 25.64%. Notably, the mean values across pond, irrigation canal, and river samples did not display a significant disparity ($P=0.48$).

In Table 2, *B. pfeifferi* was notably prevalent at the Luvungi site, making up 27.5% of the sample, while *M. tuberculata* was predominantly found at the Kiliba site, representing 25.64%. Luvungi boasted the highest snail abundance, with 46.31% of individuals across seven species, followed by Kiliba with 36.44% of specimens spanning nine species. Sange recorded the lowest snail count at 17.25%, encompassing five species. Mean values indicated no significant difference ($P=0.66$) in total snail captures among the Luvungi, Sange, and Kiliba sites.

Table 3 reveals that the diversity indices of Simpson and

Shannon-Wiener were notably elevated in the Irrigation canals, whereas the Fisher's alpha diversity index was higher in the ponds. When comparing the biological diversity across these three freshwater habitats using the Simpson, Shannon-Wiener, and Fisher's alpha indices, no significant difference was observed ($P=0.95$).

Relationship Between the Physicochemical Factors and the Snail Species

The CCA depicted four water properties that exhibited correlations with the benthic molluscan species across the sampling sites (refer to Figure 2). The initial two CCA axes accounted for 53.67% and 28.56% of the variance in benthic snail distribution, with eigenvalues of 0.8939 and 0.4757, respectively. The results indicated positive correlations between *Bulinus forskalii*, *Biomphalaria* sp., and *Bulinus globosus* with pH. Conversely, *Melanoides* sp. showed a positive association with electrical conductivity. Additionally, *Bellamya* sp. displayed a negative correlation with DO.

Prevalence Of Schistosomiasis in the Ruzizi Congolese Plain

Figure 3 illustrates that the incidences of schistosomiasis outbreaks in the village of Luvungi were notably high, with significant peaks recorded in 2010 and 2011. This

Table 1. Checklist of the Snails Collected in the Freshwater Biotopes of the Ruzizi Congolese Plain

Species	Pond	Irrigation Canal	River	Total (%)
<i>Pila ovata</i> Olivier, 1808	2.23	8.53	6.3	17.06
<i>Bellamya</i> sp.	0	0.03	0	0.03
<i>Melanoides</i> sp.	0.05	18.81	0.03	18.89
<i>Melanoides tuberculata</i> Müller, 1774	8.29	12.17	5.18	25.64
<i>Lymnaea natalensis</i> Krauss, 1848	1.14	1.86	0.19	3.19
<i>Lymnaea</i> sp.	0	0.05	0	0.05
<i>Bulinus forskalii</i> Ehrenberg, 1831	0.05	0	0	0.05
<i>Bulinus globosus</i> Morelet, 1868	1.41	0.03	0	1.44
<i>Bulinus tropicus</i> Krauss, 1848	2.84	0	0	2.84
<i>Biomphalaria</i> sp.	0.03	0	0.03	0.06
<i>Biomphalaria pfeifferi</i> Krauss, 1848	30.64	0.11	0	30.75
Total (%)	46.68	41.59	11.73	100

Table 2. The Total Relative Abundance of the Snail Species Captured at the Studied Sites

Species	Kiliba	Luvungi	Sange	Total (%)
<i>Pila ovata</i> Olivier, 1808	9.22	7.84	0	17.06
<i>Bellamya</i> sp.	0.03	0	0	0.03
<i>Melanoides</i> sp.	0.19	8.95	9.75	18.89
<i>Melanoides tuberculata</i> Müller, 1774	25.64	0	0	25.64
<i>Lymnaea natalensis</i> Krauss, 1848	0.21	0.43	2.55	3.19
<i>Lymnaea</i> sp.	0	0.05	0	0.05
<i>Bulinus forskalii</i> Ehrenberg, 1831	0.05	0	0	0.05
<i>Bulinus globosus</i> Morelet, 1868	0.72	0.72	0	1.44
<i>Bulinus tropicus</i> Krauss, 1848	0	0.82	2.02	2.84
<i>Biomphalaria</i> sp.	0.03	0	0.03	0.06
<i>Biomphalaria pfeifferi</i> Krauss, 1848	0.35	27.5	2.9	30.75
Total (%)	36.44	46.31	17.25	100

Table 3. Biological Diversity Indices in the Freshwater Habitats

Diversity Indices	Freshwater Habitats		
	Pond	Irrigation Canal	River
Simpson (1-D)	0.53	0.67	0.52
Shannon-Wiener (H')	1.12	1.22	0.79
Fisher's α	1.24	1.10	0.79

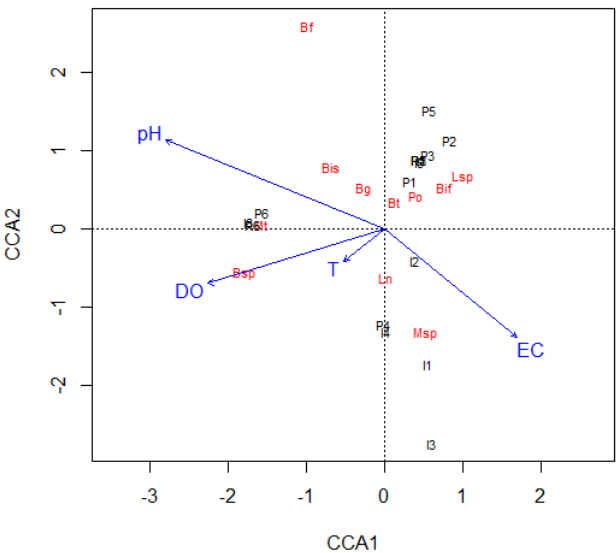


Figure 2. CCA Expressing the Physicochemical Parameters Effect on Mollusc Communities; Dissolved Oxygen (DO), Temperature (T), Electrical Conductivity (EC), *Pila ovata* (Po), *Bellamya* sp. (Bsp), *Melanoides* sp. (Msp), *Melanoides tuberculata* (Mt), *Lymnaea natalensis* (Ln), *Lymnaea* sp. (Lsp), *Bulinus forskalii* (Bf), *Bulinus globosus* (Bg), *Bulinus tropicus* (Bt), *Biomphalaria* sp. (Bis), *Biomphalaria pfeifferi* (Bif), Ponds (P1-P6), Irrigation Canals (I1-I6)

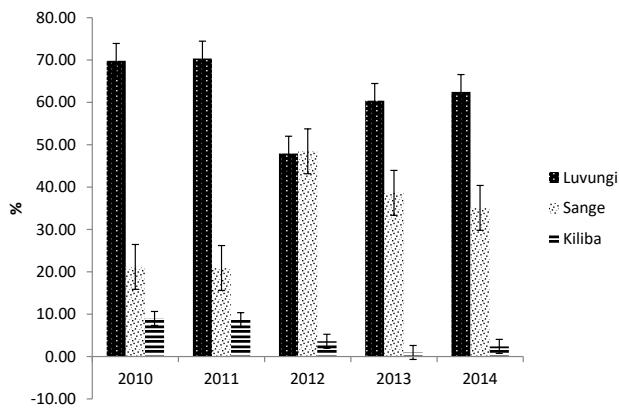


Figure 3. Outbreaks of infection of schistosomiasis

village harbored considerable populations of *B. pfeifferi* infested with trematode parasites. Conversely, there was a decline in cases observed in the Sange village following a rise from 2011 to 2012.

Figure 4 demonstrates a significant positive Pearson's correlation between species abundance and schistosomiasis. In regions where *B. pfeifferi* was abundant, the infestation rate of schistosomiasis was notably high.

The inventory of freshwater malacological fauna revealed a total of 11 species belonging to the Gastropoda class, with *Bulinus tropicus* and *Bellamya* sp. being newly documented in the study area. This discovery aligns with the broader observations of malacological fauna in the freshwaters of the Ruzizi Congolese plain. Among the 11 species identified in the Ruzizi Congolese plain, only *B. pfeifferi* exhibited high abundance in the freshwaters and was exclusively found in ponds and irrigation canals. The presence of invasive species in our sampling sites notably impacted the local molluscan fauna, exemplified by the competitive effectiveness of *M. tuberculata* against medically significant planorbids.^{9,21}

Our findings largely coincide with those reported by Muhigwa et al on freshwater snails in Kiliba, encompassing species like *Pila ovata*, *Lymnaea natalensis*, *B. pfeifferi*, *Biomphalaria* sp., *M. tuberculata*, *Physa acuta*, *Bulinus truncatus trigonus*, and *Planorbis* sp.¹²

The molluscan gastropods documented in the surveyed sites of Kiliba, Luberizi, and Sange from 1979 to 1980 included *Lymnaea natalensis*, *B. pfeifferi*, *Biomphalaria sudanica*, *Bulinus forskalii*, *Bulinus truncatus*, *Gyraulus costulatus*, and *Pila ovata*. Meanwhile, Safari et al identified five genera of gastropods, namely *Biomphalaria*, *Bulinus*, *Lymnaea*, *Melanoides*, and *Pila*, in the natural ponds of the Ruzizi plain.²²

Gryseels¹³ documented the presence of various snail species, including *Pila ovata*, *Lymnaea natalensis*, *B. pfeifferi*, and others, in the Ruzizi valley of Burundi. They found that *B. pfeifferi* inhabited all freshwater habitats except rivers and was the exclusive intermediate host for *S. mansoni* in the area.

A survey of freshwater snails in Ituri (DR Congo)

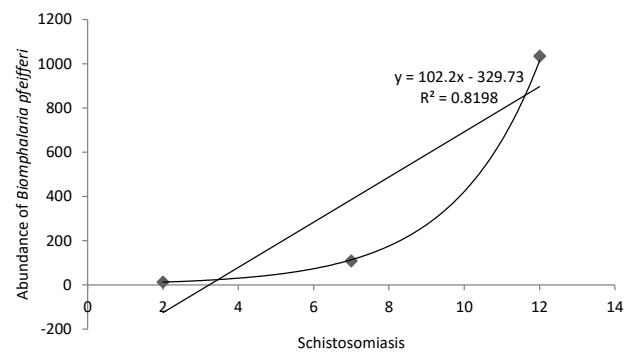


Figure 4. Importance of *Biomphalaria pfeifferi* in the Schistosomiasis Stemmed From *Schistosoma mansoni*

from 1986 to 1988 listed several species, with similarities to findings by Chartier et al²³ regarding *B. pfeifferi* distribution in Ituri.

Research by Schultheiß et al in Eastern Congo identified various Gastropoda species and noted that *B. pfeifferi* was found in irrigation channels.¹¹

Several studies, including those by De Clercq,³ Day and De Moor,⁵ and others,²⁴⁻²⁶ highlighted *B. pfeifferi* as the primary intermediate host for *S. mansoni* in tropical Africa. *Bulinus forskalii* and *B. globosus* were identified as hosts for *S. intercalatum* and *Schistosoma haematobium*, respectively, while *Lymnaea natalensis* serves as a host for *Fasciola gigantica* and *Fasciola hepatica*.

The prevalence of schistosomiasis was evaluated in three selected sites, documenting outbreaks and gathering medical statistics from 2010 to 2014. The highest prevalence was found in Luvungi, where *B. pfeifferi* populations were most heavily infested with trematode parasites. *S. mansoni* was particularly endemic in irrigated rice farming areas of the Ruzizi plain.²⁷ Conditions conducive to *B. pfeifferi* proliferation and infestation included ample light, abundant aquatic vegetation, stagnant water, and high levels of human waste.^{28, 29}

In 2015, a significant positive correlation ($R^2 = 82\%$) was observed between *B. pfeifferi* abundance and schistosomiasis prevalence. Baluku¹⁴ found high schistosomiasis prevalence in Kiliba during 1979 and 1980, correlating with a high infestation rate of *B. pfeifferi* in the same village.

Conclusion

This study identified *B. pfeifferi* as the predominant gastropod species collected from ponds, irrigation channels, and rivers in Luvungi, Sange, and Kiliba villages within the Ruzizi Congolese plain. Our data indicates that although schistosomiasis was widespread in the area, its prevalence was particularly high in Luvungi. In this region, only *B. pfeifferi* was found to shed *S. mansoni* cercariae.

Based on our findings, we recommend conducting extended, periodic monitoring of mollusc populations, potentially spanning up to three years, to observe changes in both mollusc populations and infection rates.

Furthermore, further research is necessary to determine the specific timing of *B. pfeifferi* cercariae release into the water, which could aid in developing a schistosomiasis control strategy.

Competing Interests

The author declares no conflicts of interest relevant to the content of this paper.

Ethical Approval

This article did not involve any ethical considerations.

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