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Chapter

Seasonal Variations of Densities of *Biomphalaria pfeifferi*, the Intermediate Host of *Schistosoma mansoni* Parasite at the North of Senegal

Sidy Bakhoum, Christopher J.E. Haggerty, Cheikh Tidiane Ba, Nicolas Jouanard, Gilles Riveau and Jason Robert Rohr

Abstract

Schistosomiasis is becoming more persistent because of the widespread distribution of intermediate host snails in several regions of Africa, including Senegal. The intermediate snail host of the human intestinal schistosome is *Biomphalaria pfeifferi* and is permanently present in northern Senegal because of the presence of the abundant freshwater habitat throughout the year. Here, we observed the seasonal variation in *B. pfeifferi* abundance in the Saint-louis region at the North of Senegal in West Africa. We performed snail and environmental parameter sampling across two different seasons described for Senegal: a dry season that runs roughly from mid-October to mid-June and a rainy season that spans approximately from late June to early October. We also split the dry season into two categories representing periods of time when water temperatures were either decreasing (dry1) or increasing (dry2). We used regression analyses to model snail density across the seasons and investigated which environmental variables influenced snail abundance. Results suggested that snails were more abundant and peaked during the rainy season, which lowest abundances during the dry season when temperatures were declining. The above seasonal variations of snail density were positively linked to the environmental drivers including periphyton (food resource for snails), aquatic vegetation abundance, water temperature and dissolved oxygen and negatively to both pH and water conductivity. Our findings may be useful for snail control efforts by targeting specific periods and/or site conditions when snail abundances are greatest.

Keywords: schistosomiasis, intermediate hosts, snails, seasonal variation, *Biomphalaria pfeifferi*, *Schistosoma mansoni*

1. Introduction

To completely understand many infectious disease systems, it will become increasingly important to understand how seasonality affects multiple processes, including (but not limited to): host behavior, reproduction, survival in the environment [1]. Seasonal variations in temperature, rainfall and resource availability are ubiquitous and can exert strong pressures on host population dynamics.

Infectious diseases provide some of the best-studied examples of the role of seasonality in shaping population fluctuations [1]. Predicting disease dynamics requires an understanding of the traits of hosts across seasons.

Schistosomiasis remains a significant health burden in many areas of the world [2]. Intermediate hosts snails of human schistosomes release parasites that cause human schistosomiasis, are strongly driven by environmental factors [3–5]. The transformation of ecosystems in the river delta Senegal has created favorable biotopes to the development of intermediate host snails of human schistosomes [3], and schistosomiasis rates have increased from historic levels. Thus, limiting or controlling snail populations is an important step in disease control. Yet, consideration of the relative influence of seasonality on the environmental factors that influence snail host populations remains needed.

In this study, we investigated how snail host abundance varies at water access sites in North Senegal, West Africa, a location where human schistosomiasis prevalence is among the highest globally. We conducted our study in Senegal River at four sites used for water access in three villages. We performed biweekly monitoring of *Biomphalaria pfeifferi* across two different seasons in Senegal: a dry season that runs roughly from mid-October to mid-June and a rainy season spanning approximately from late June to early October. Further, we sampled two periods of water temperature T (°C): Dry_1 (periods of decreasing temperature ($29.4 \geq T(Dry_1) \geq 15.4$) and Dry_2 (periods of increasing temperature ($15.5 < T(Dry_2) \leq 32.5$)). We conducted a total of $n = 21$ visits to survey density of hosts and the environmental parameters in the field (as described below) for one year from October 2019 to October 2020.

2. Methods

2.1 Study sites

We sampled intermediate hosts biweekly at four water access points across three villages in Senegal, West Africa: Kaban (KA: 16° 3.338 N - 16°24.133 O) Minguegne (ME 1: 16°01.055' N - 16°21.397' O and ME2:16°01.090' N - 16°21.369 O) and

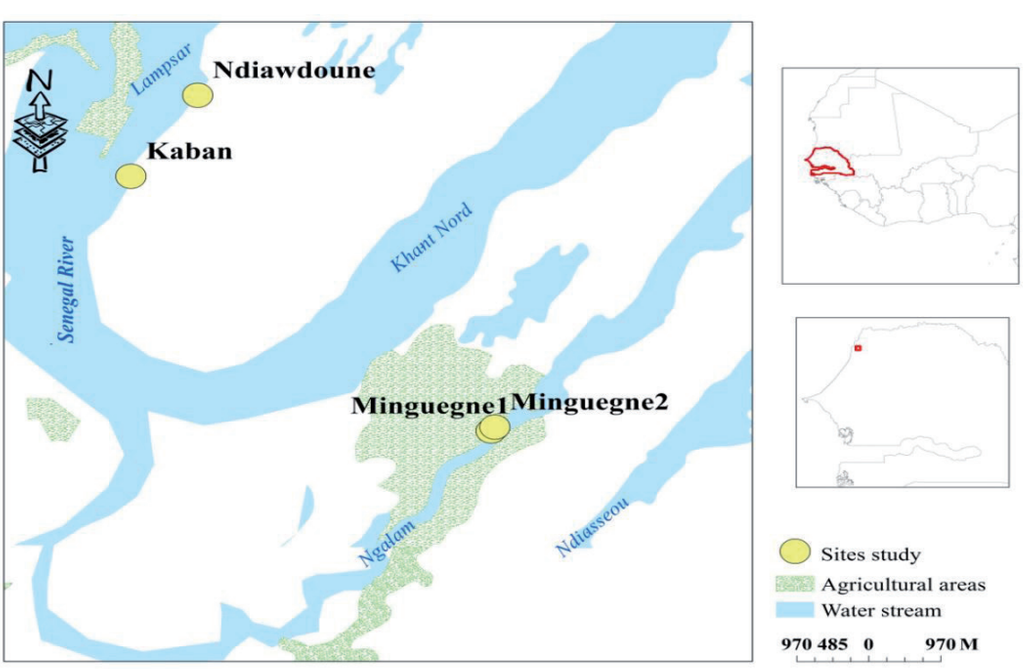


Figure 1.
Location of the study sites along the Senegal River in West Africa.

Ndiawdoune (NW: 16°4.075' N - 16°23.635' O). Kaban village is bordered by the Senegal river, Minguegne is bordered to the Ngalam outlet Senegal River in the "Trois marigots" zone and Ndiawdoune is bordered by the Lampsar River (**Figure 1**).

2.2 Environmental factors driving host snail abundance

We recorded dissolved oxygen (DO), pH, water conductivity and water temperature using a YSI Professional Plus handheld multiparameter meter. We also recorded periphyton fluorescence using an Aquapen AP 100-C handheld flourometer. Periphyton was collected from a study site during snail sampling (see below) by wading into the water and cutting a stem of *Typha* spp. vegetation at the water surface and again at a depth of 10 cm below the water. The 10 cm section of Typha was taken to the lab and its surface scrubbed using a toothbrush. We then washed all algae off the Typha and brush using deionized water into a 50 mL falcon tube and filled all samples to a standardized volume of 50 mL. Chlorphyll a was then quantified by filling a 1 mL cuvette and recording Ft values using the Aquapen. We recorded the surface area of *Typha* that was sampled and used it to standardize the periphyton flouresence values based on sampling area.

2.3 Snail sampling

We used the snail sampling method described in [6]. At each water access point, we conducted 10 1-m sweeps with a 2.5-mm mesh aquatic dipnet at random sampling points. Any aquatic plants in the dipnet were placed into a wash pale with water, shaken vigorously to remove snails, and examined for any attached snails before weighing the vegetation mass using a spring scale. We recorded the number of *Biomphalaria pfeiferii* snails (**Figure 2**) captured per sweep.

2.4 Statistical analysis

We conducted our data analysis using R software version string *R version 4.0.5 (2021-2103-31)*. We applied a forest plot in the *Forest model* package to a linear model to assess the variation of snail's density across the seasons. In forest plots, a line of no effect (at 0) marks the point where there is no clear difference between the variables. If the 95% confidence intervals (CIs) do not cross the line of no effect, then the result is significant (p-value <0.05). To elucidate actions between



Figure 2.
Biomphalaria pfeifferi *snail species of the current study. This is the only known intermediate host of* Schistosoma mansoni, *the parasite of the intestinal schistosomiasis in Senegal.*

environmental parameters and hosts density, we applied Pearson's correlation, which gives a measure of the strength of a linear association between two variables. We also determined the *p-value* or probability that we would have found the current result if the correlation coefficient *r* were in fact zero (null hypothesis). If this probability is lower than the conventional 5% ($p < 0.05$) the correlation coefficient is called statistically significant. We used linear model in *ggplot2* package that utilizes a *lm* function to assess the significance of linear predictor-response relationships between hosts and environmental drivers.

3. Results

3.1 Seasonal variations and environmental parameters driving the densities of the snail's hosts

We collected a total *895 Biomphalaria pfeifferi*, including 83 in Dry1 (9.27%), 192 in Dry2 (21.45%), and 620 in rainy (69.28%) seasons (**Table 1**). A factor for season in our forest plot analyses was a significant predictor of snail abundance (**Table 2**, *p-value* = 0.003), with both greater mean of total density (*N*) and maximum snail density per visit occurring in the rainy season (**Table 1**). In contrast, all measures of snail abundance were lowest during Dry1 season, whereas a medium density occurred in Dry2 season (**Figure 3**). Our forest plot analyses suggested that snail abundance in the rainy season was significantly different from season Dry1, whereas season Dry2 had lower but not significantly lower snail abundance compared to the rainy season (**Figure 2**; *p-value* = 0.181).

During Dry1 season, when snail abundance was lowest, no environmental parameters had a significant association to *Biomphalaria* abundance (**Table 1**;

		Mean	Max
Seasons	Dry1 (*N = 83*)	3.458	18.000
	Dry2 (*N = 192*)	9.6	33.0
	Rainy (*N = 620*)	15.5	87.0
Total *(N = 895)*		10.65	87.0

Table 1.
Total, mean and maximum of the total and seasonal densities of Biomphalaria pfeifferi.

Variable		N	Estimate	p
Seasons	Dry1	24		Reference
	Dry2	20		6.14 (-2.92, 15.20) 0.181
	Rainy	40		12.04 (4.32, 19.77) 0.003

0 5 10 15

Table 2.
Forest plot analyses of Biomphalaria pfeifferi *abundance across seasons.*

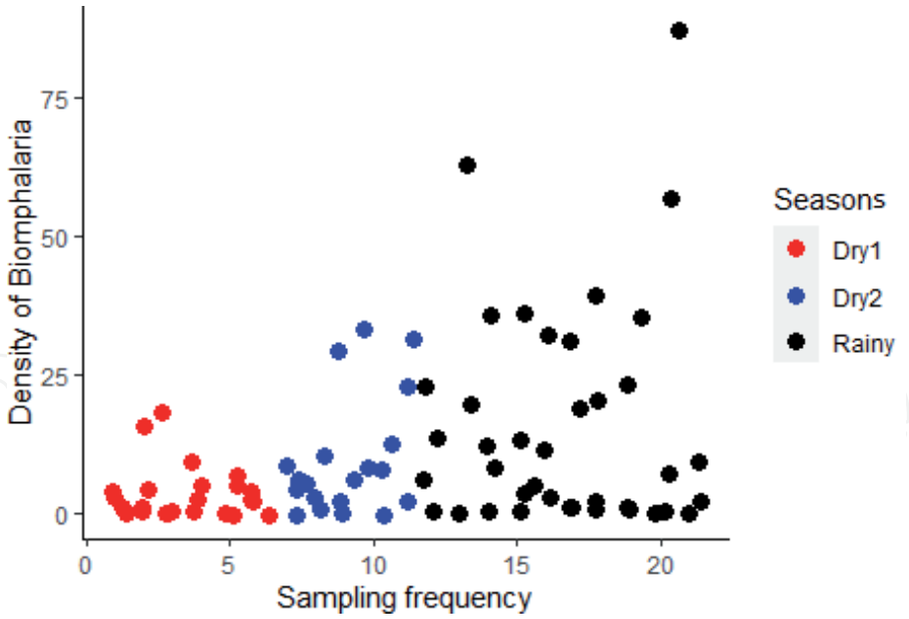


Figure 3.
Plot showing the distribution of Biomphalaria pfeifferi *across the seasons. The greatest density was collected during the rainy season (with a peak >75) while the weakest was found during the Dry1 season.*

Environmental parameters	Density of *Biomphalaria pfeifferi*		
	Dry1	**Dry2**	**Rainy**
Temperature (°C)	0.991	0.04*	0.549
Conductivity (μS/cm)	0.111	0.02*	0.08
DO (mg/l)	0.447	0.006**	0.04*
Periphyton	0.431	< 0.001***	0.003**
Mass vegetation (g)	0.452	0.007**	0.05
pH	0.429	0.03*	0.388

**P ≤ 0.05*
***P ≤ 0.01*
****P ≤ 0.001*
If P > 0.05, there is no significance (no asterisk).

Table 3.
Linear regressions between environmental factors and density of Biomphalaria pfeifferi for each season of snail sampling.

Figure 3; *p-value* > 0.05). In contrast, several environmental factors were significantly correlated to the snail host abundance in season Dry_2 (**Table 3**). Conductivity and pH were negatively ($r < 0$) and significantly associated to hosts abundance whereas other drivers are positively ($r > 0$) correlated in season Dry_2. The maximum water temperature (32.5° C) was obtained during rainy season. Water temperature was positively related to snail abundance, exerted a significant effect only in the season Dry_2 (**Table 3** and **Figure 3**). There was no significant effect of temperature on snail densities in the rainy season (p-value = 0.549). Only the dissolved oxygen (p-value = 0.04) and periphyton (p-value = 0.003) were significantly correlated to the density of hosts in the rainy season. Periphyton was a significant positive predictor of snail abundances in both the Dry_2 and rainy seasons (**Figure 4**).

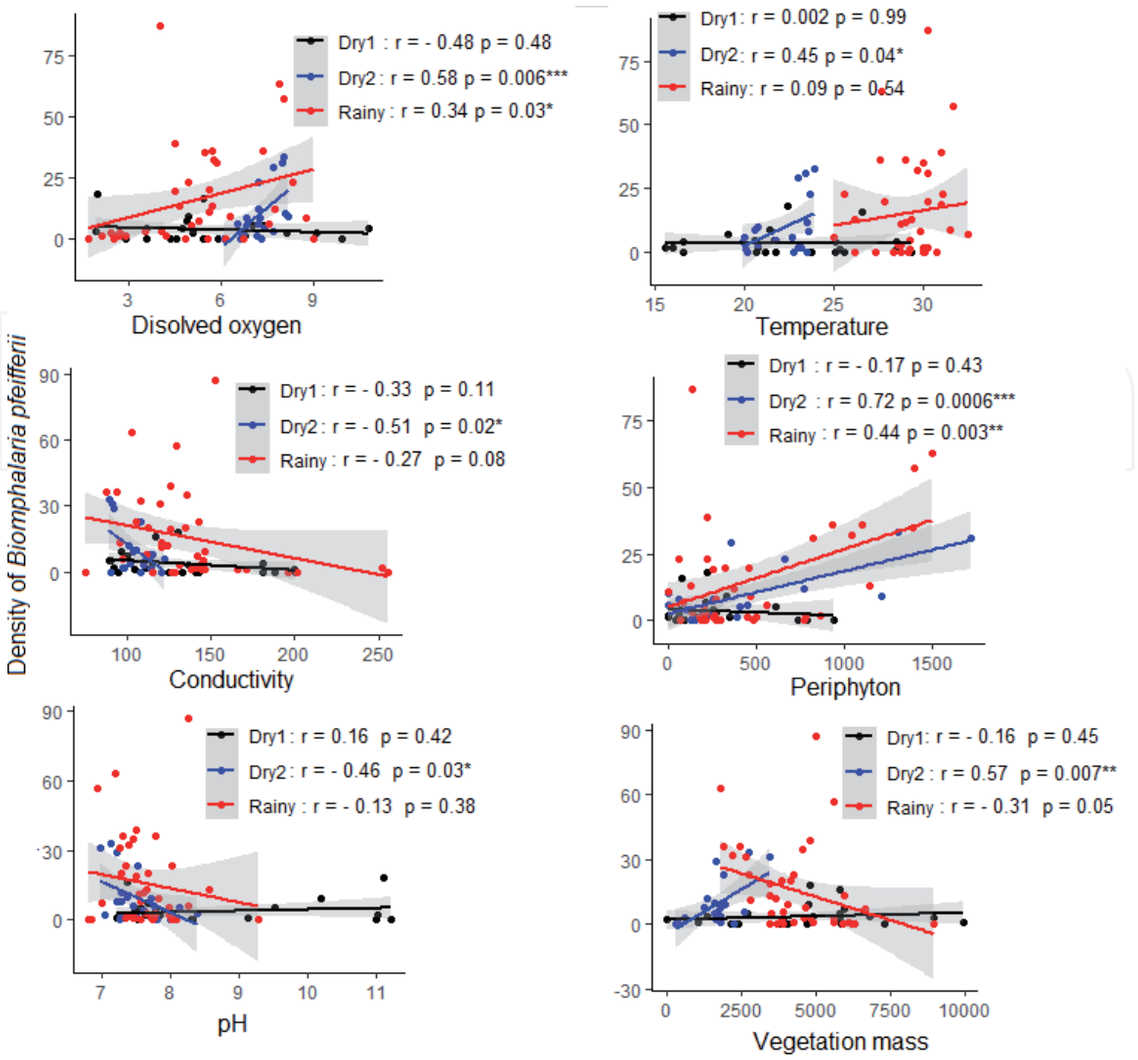


Figure 4.
Linear model assessing the significance of predictor-response relationships between environmental parameters and Biomphalaria pfeifferi *densities across the seasons.*

4. Discussion

4.1 Seasonal variations and environmental parameters driving the densities of snail hosts

Ecosystem changes in the delta of the Senegal River has created favorable biotopes to the development of intermediate host snails of human schistosomes, which may have included particular the physicochemical conditions favorable to their abundance [3]. Permanent freshwater is associated with aquatic floating vegetation such as *Ceratophyllum sp* and *Ludwigia sp* that acts as beneficial snails habitat in several areas in the Delta of Senegal River. The presence of the intermediate host *Biomphalaria pfeifferi* was previously described in several studies in the study area [3, 6–8]. In this area, *Biomphalaria pfeifferi* was the second most abundant intermediate host of human schistosomiasis after *Bulinus truncatus* [3]. Our study highlights that *B. pfeifferi* abundance is lowest during a Dry_1 season likely because of lower periphyton and water temperature than in the rainy season. Lower water temperature during Dry_1 season can limit snail growth and reproduction. For example, *Biomphalaria glabrata*, a neotropical species that is another intermediate host for *S. mansoni*, generally avoids thermal extremes and

prefers temperatures from 27° to 32° C [9]. The greater density of *Biomphalaria* during periods of increasing temperature (19.9° to 32.5°C) during Dry_2 and Rainy is increasing abundance of periphyton that acts as snail food, as well as periods of higher dissolved oxygen, temperature and aquatic vegetation that acts as snail habitat. In contrast, during this same period, negative environmental influences on snail abundance including pH and conductivity are declining. Aquatic vegetation provides both habitat and food resources to snail hosts and is itself positively associated to the snail abundance [6], while high conductivity and pH impact negatively snails' intermediate hosts of human schistosome [3]. Although 55.1% of total vegetation mass was collected in the rainy season, vegetation was only a significant predictor of snails in the Dry2 season (10.9%). *Biomphalaria* species prefer areas of clear water on sandy and gravel bottoms, with stagnant water or with a very light current, with sometimes abundant aquatic vegetation [9]. Snail abundance was most associated with oxygen during the Dry_2 season, and *Biomphalaria pfeifferi* is known to be positively associated with dissolved oxygen [3], whereas pH in the range 5.0 to 7.5 is likely a weaker determinant of snails [9]. We suspect that snail density was not significantly correlated to water temperature because of every high temperature (32.5° C) we found during the rainy season. This driver could limit reproduction and other physiological functions or be lethal to the snail survival. *Biomphalaria pfeifferi* does best under warm stable conditions [10]. Thus, future studies should also consider non-linear relationships in their analyses. We found that the month of August was the most important for snail control because densities of host during the rainy season were fueled by ideal environmental conditions in freshwater (i.e: resource food availability that may increase host reproduction). During this period, rainwater transports organic wastes, fertilizer and pesticides from agricultural areas to adjacent waters. Such agrochemical runoff may affect the development aquatic vegetation and periphyton and pesticide pollution is a major driver in increasing the occurrence of host snails [11, 12]. Mesocosm studies support the assertion that fertilizer, herbicide, and insecticide, individually and as mixtures may be increasing the algae snails eat in the rainy season [13]. Moreover, such chemicals can decrease the densities of snail predators. Our findings may be context dependent to the Senegal River as seasonality may favor the dry season months in other contexts [14].

5. Conclusion

Given the widespread distribution of *Biomphalaria pfeifferi* in the Senegal River, understanding the seasonal variation and the principal drivers of snail abundance is important for local snails control to prevent upcoming human risk of *Schistosoma mansoni* infection. Our findings support that the rainy season is significantly associated (p-*value* = 0.003) with the abundance of intermediate hosts because of highly favorable environmental conditions for periphyton (snail food) and dissolved oxygen levels required by *B. pfiefferi*. Conditions were less ideal in the Dry_2 season than the rainy season, and, thus, more environmental parameters were significant for this period than in the rainy season. In contrast, environmental conditions were so poor in the Dry_1 season that few snails occurred and significant trends were absent. Our findings on the seasonal fluctuations of snail hosts are useful for targeting a snail control program during a time of year when it may be most effective to eliminate or reduce the vectors of schistosomiasis.

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