Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 – 6131
Vol. 28(5): 755 – 767 (2024)
www.ejabf.journals.ekb.eg

Schistosomiasis and Geographical Distribution of Aquatic Mollusks and Disease Vectors in the Gharb Plain (Morocco)

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ARTICLE INFO

Article History:
Received: Aug. 13, 2024
Accepted: Sept. 2, 2024
Online: Sep. 18, 2024

Keywords:
Schistosomiasis,
Mollusks,
Gharb plain,
Morocco

ABSTRACT

This research work presents the malacological and schistosomiasis status in the Gharb, one of the large plains in northwestern Morocco, just before its redevelopment following the commissioning of a new and large dam, El Wahda. The results show that no danger was posed to humans by the populations of *Planorbis metidjensis* (Forbes, 1838) during the current study. In addition, an overlap of the distribution of schistosome infestation was monitored and the intermediate hosts raise several issues, notably: (1) the problem of the external or local origin of schistosomiasis in certain schistosomiasis foci in the Gharb plain; (2) a change, in time and space, in the geographical distribution of the intermediate host foci is possible; (3) a change in the infestation rate of the populations of the intermediate host is also possible; (4) *Physa acuta* (Draparnaud, 1805), the species whose presence is constant in schistosomiasis foci and which is experimentally infectable by *Schistosoma haematobium* (Bilharz, 1852) in the literature, may likely constitute a third vector of schistosomiasis in Morocco.

INTRODUCTION

Poor populations in temperate or warm climate countries suffer from parasitic infections, particularly neglected tropical diseases such as schistosomiasis, malaria, leishmaniasis, and trypanosomiasis **(Mostafa & Atef, 2024)**. Schistosomiasis, the second most common parasitic disease in the world, is a widespread chronic tropical disease posing a public health problem in several developing countries **(Adenowo *et al.*, 2015; El-Karim, 2022)**. Victims are infected through contact with the larvae of the parasite, which are released by intermediate host mollusks in the aquatic environments of warm countries. Once the parasite penetrates the skin, it migrates into the human body and localizes in a suitable organ such as the urinary system, liver, or hepatic system. Thus, depending on the final location of the adult parasites in the patient's organs, there are several types of schistosomiasis: urogenital, hepatic, and intestinal (or rectal) schistosomiasis. High parasitic loads lead to lesions in the affected organ. Numerous mollusks, all gastropods, serve as intermediate hosts for the parasitic disease (Table. 1).

Table 1. Parasite species and geographical distribution of schistosomiasis **(Kokaliaris *et al*., 2022)**

Type of bilharzia	Species	Geographical distribution
Intestinal schistosomiasis	*Schistosoma mansoni*	Africa, the Middle East, the Caribbean, Brazil, Venezuela and Suriname
	Schistosoma mekongi	Several districts of Cambodia and the Lao People's Democratic Republic
Sino-Japanese spleno-hepatic schistosomiasis	*Schistosoma japonicum*	China, Indonesia, the Philippines
Urogenital and digestive system schistosomiasis	*Schistosoma guineensis* and Related *S. intercalatum*	Rain forest areas of central Africa
Urogenital schistosomiasis	*Schistosoma haematobium*	Africa, the Middle East, Corsica (France)

Estimates show that at least 251.4 million people needed preventive treatment for schistosomiasis in 2021, of which more than 75.3 million people are estimated to have been treated **(Kokaliaris *et al*., 2022)**. Urogenital schistosomiasis, caused by *Schistosoma haematobium*, is the most common form of human schistosomiasis **(Alphonse, 2022)**. It is prevalent in 54 countries in Africa and West Asia **(Ferandel, 2001)**. It was reported that at least 90% of those requiring treatment for schistosomiasis live in Africa **(Kokaliaris *et al*., 2022)**.

Regarding the fight against schistosomiasis, two main methods are used either separately or in combination: one method is based on the treatment or good hygiene behavior of the patient or the person exposed to the disease, and the other method is based on controlling the intermediate vector **(World Health Organization, 2004)**. Thus, as indicated by **Jobin (2020)**, the methods included biological control of schistosome transmission, control of snails through general application of Bayluscide (niclosamide), and control of snails through focal application of Bayluscide. These constitute an integrated strategy for combating schistosomiasis.

Apart from praziquantel, the primary drug used against bilharzia in many countries, the development of a therapeutic arsenal against schistosomiasis has contributed globally to reducing the endemicity of the disease **(Savioli *et al*., 2017)**. This reduction in the endemicity of the disease is attributed to the eradication and/or treatment of intermediate vector habitats using molluscicides **(Belkacemi & Jana, 2006; Poda, 2007)**. In some cases, it involves modifying irrigation channels to increase water speed to prevent the establishment of intermediate vectors **(Cecchi *et al*., 2007)**. However, the reasons for the disappearance of bilharzia in a region are not always easy to explain. The epidemiological case of Gharb, a vast plain in northwestern Morocco, is one such instance.

Thus, in this study, we aimed to contribute to understanding the key characteristics of the epidemiological evolution of schistosomiasis in Morocco, focusing on its occurrence in the Gharb plain.

MATERIALS AND METHODS

A- Study area

The Gharb plain (Fig. 1) is located in the northwest of Morocco, 40km north of Rabat. Its total area is estimated at 400km². The climate is of the Mediterranean type, with a sub-humid bioclimatic layer to the west and semi-arid to the east. Moreover, the surface hydrological network is very diverse.

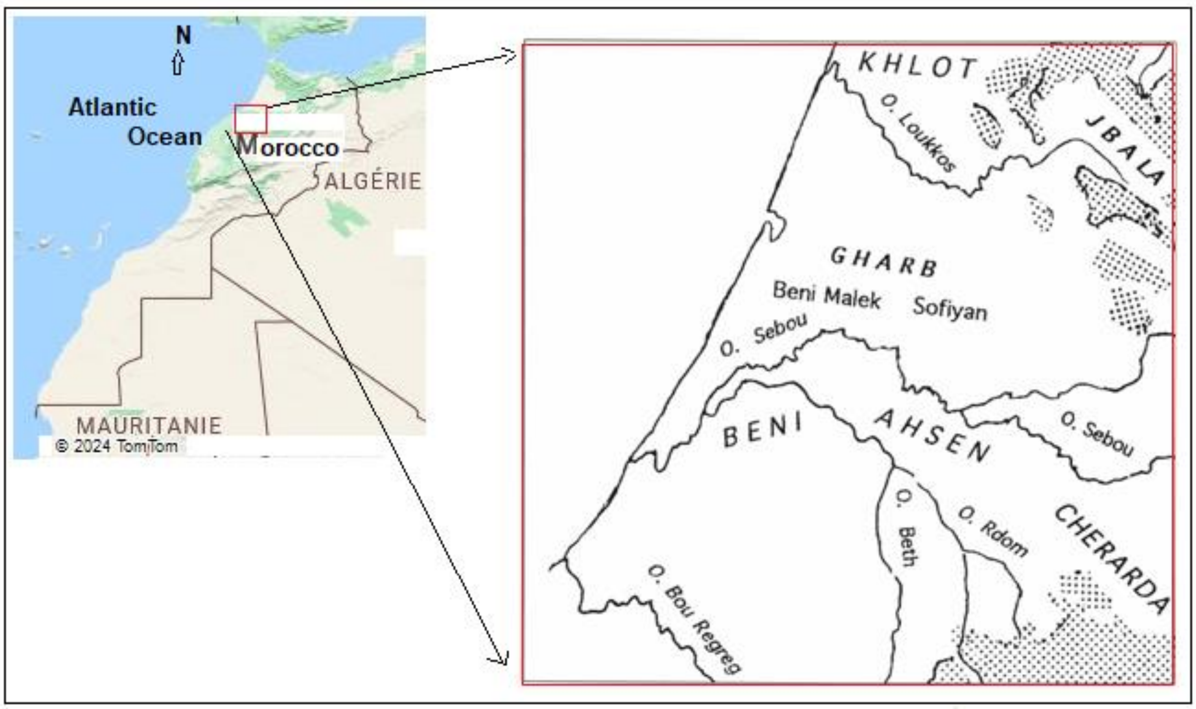

Fig. 1. Geographical location of the Gharb plain

B. Malacological sampling

During the period 2018-2023, we conducted seasonal qualitative sampling of the malacological fauna at 61 stations, the distribution of which covers the different types of surface accumulations in the plain. The selection of stations took into account available malacological data **(Hadji, 1980; Kharboua, 1988; Maqboul, 1996; Fadli, 2003)**, the proximity of the stations to human settlements, and the spatio-temporal variability of the water bodies.

Moreover, the significant seasonal fluctuations in the abundance of mollusks, the size variations among individuals, and the complexity of their microhabitats **(Mouthon, 1980)** led us to combine various methods of sampling the malacological community.

- Sampling by seine net

This type of operation, also carried out by **Aguesse (1961)** to sample the aquatic zoocenosis, is used in the sampling of the malacofauna of shallow water accumulations.

- Sampling by metal dredge

This is the sampling method that seemed most suitable for collecting mollusks from pits and relatively deep canals. The dredge used is equipped with a mosquito net with a mesh size of 0.8mm and a metal frame that allows for scraping the edges of these types of biotopes. This same technique was used by **Khallayoune and Laayerant (1992)**, **Tsafack *et al*. (1995)**, **Fadli (2003)**, **Sturm *et al*. (2006)**, and **Anderson *et al*. (2013)**.

- Sampling with a Surber net

At the flowing water stations, we used a square sieve (filter that also serves as a screen) measuring 25cm on each side and equipped with a mosquito net with 0.8mm mesh openings. The sampling operation involves scraping the bottom over a well-defined large area in front of this filter; the malacological fauna caught by the sieve is collected.

- Hand collection (or Visual hunting)

This method, which involves searching for individuals on mud, on vegetation, under stones, or on any other substrate that may harbor mollusks, allows for the completion of the faunal list of collected species. This same technique has been adopted by numerous authors including **Coulibaly and Madsen (1990)**, **Ngonseu *et al*. (1992)**, and **Dillian and Bello (2020)**.

C. Parasitological survey

According to the population abundance, 50 to 90 individuals belonging to different size classes are randomly selected and subjected to the cercarial emission test, which involves exposing each individual to a light source for a duration of 24 hours **(Christensen *et al*., 1984; Théron, 2015)**. When infested, the mollusk emits the infective larvae. A total of 670 individuals of *P. metidjensis* are subjected to this test. Although this method is not very reliable, the ratio of the number of infested individuals to the number of analyzed individuals allows for an estimation of the infestation rate of the studied populations.

D. Geographical distribution of schistosomiasis hotspots

The hotspots of schistosomiasis reported in previous works and the data provided by the local authorities of the Ministry of Public Health (S.I.A.A.P) of the city of Kénitra allowed us to create a geographical distribution map of schistosomiasis hotspots in the studied plain.

RESULTS

During our study, we collected 16 species belonging to 8 families of gastropods:
Subclass: Pulmonata
Order: BASOMMATOPHORA
Family: Lymneidae
- *Lymnaea truncatula* (Müller, 1774)
- *Lymnaea peregra* (Müller, 1774)
- *Lymnaea stagnalis* (Linné, 1758)
- *Lymnaea palustris* (Müller, 1774)

Family: Planorbidae
- *Planorbis planorbis* (Linné, 1758)
- *Planorbarius metidjensis* (Forbes, 1838)
- *Gyraulus crista* (Linné, 1758)

Family: Physidae
- *Physa acuta* (Draparnaud, 1805)

Family: Ancylidae
- *Ancylus fluviatilis* (Müller, 1774)

Family: Acroloxidae
- *Acroloxus lacustris* (Linné, 1774)

Order: STYLOMMATOPHORA
Family: Succinneidae
- *Succinea debilis* (Morelet, 1859)

Subclass: Prosobranchia
Order: MONOTOCARDIA
Family: Melanoidae
- *Melanopsis praemorsa* (Linné, 1758)
- *Melanopsis costellata* (Ferussac, 1923)
- *Melanopsis scalaris* (Gassies ,1856)

Family: Hydrobiidae
- *Mercuria confusa* (Frauenfeld, 1838)

It was noted that the Lymnaeidae and Planorbidae are the two families specifically best represented. *Bulinus truncatus*, which is known as the main intermediate vector of *S. haematobium* in Morocco, is absent. Fig. (2) and Table (2) represent the geographical distribution of all the foci of schistosomiasis in the studied area.

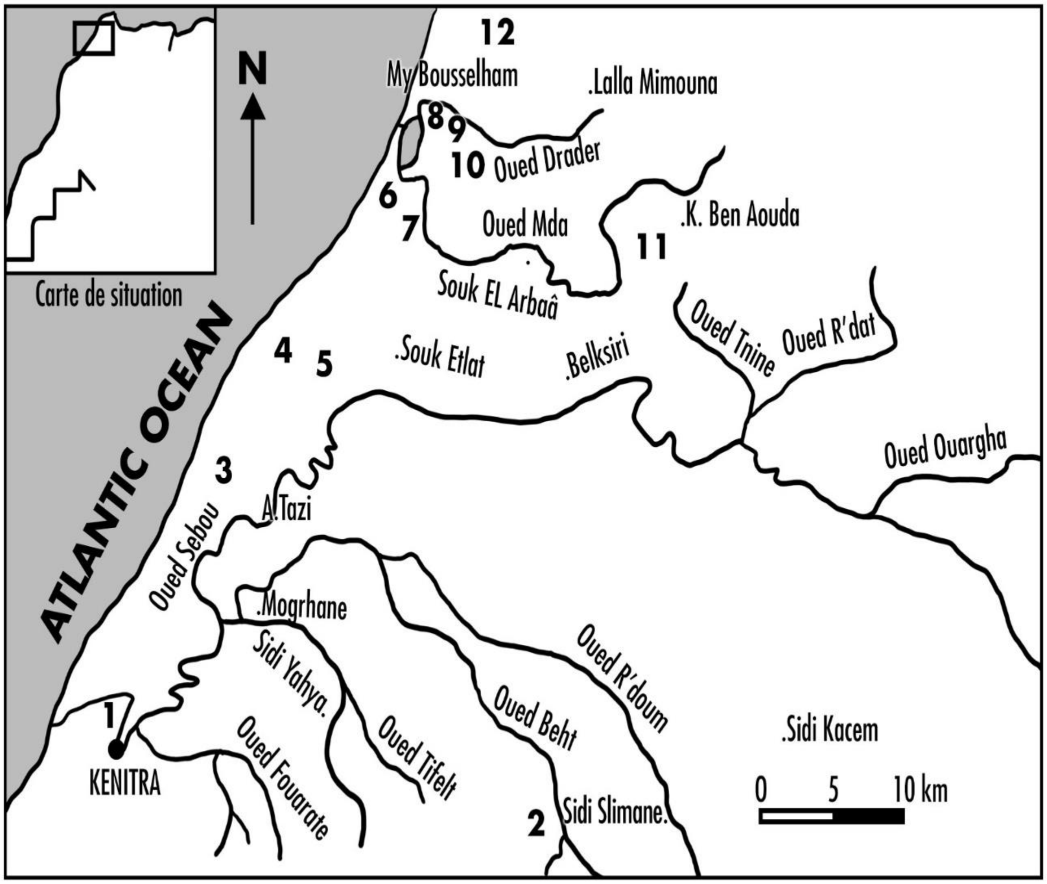

Fig. 2. Distribution of surveyed stations in the Gharb plain

Furthermore, previous studies that have addressed the epidemiological and/or malacological study of the Gharb plain show the existence of several focal points where schistosomiasis has been recognized by the S.I.A.A.P. of the city of Kenitra as an allochthonous area of schistosomiasis (Fig. 3).

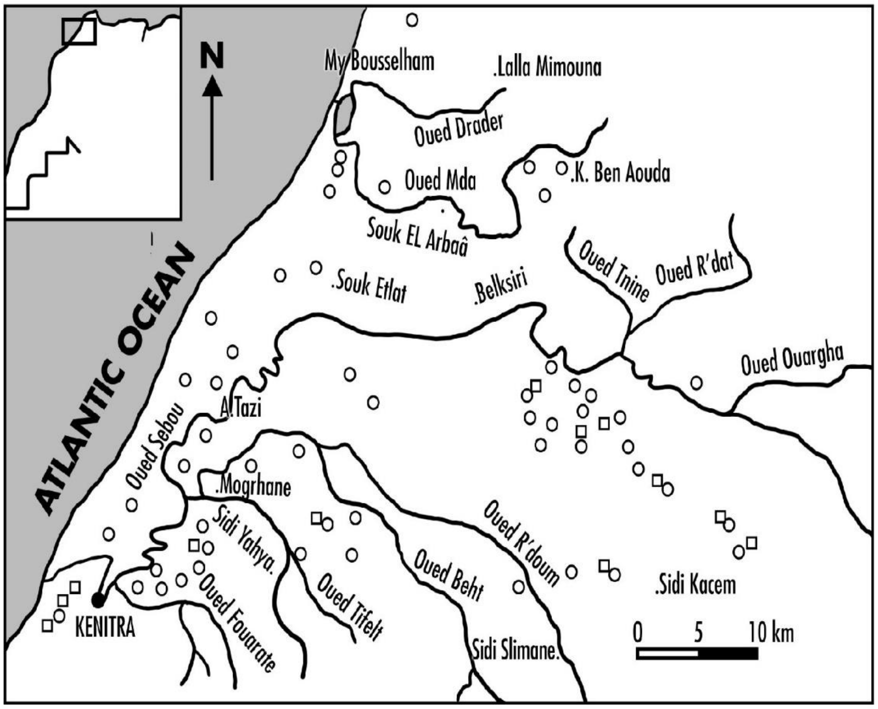

Figure 3. Geographical distrubution of *P. metidjensis* (□) *and P. acuta* (o) in the Gharb plain

Table 2. Geographic sites that have been historically reported by the S.I.A.A.P. as allochthonous bilharzia points

Geographical sites that have been historically reported by the S.A.A.P. as allochthonous bilharziasis points	Symbol	Geographical coordinates
Kenitra	**1**	34°15′N 6°35′W
Dar Bel Amri	**2**	Latitude: 34.1889, Longitude: -5.96972 34° 11′ 20″ Nord, 5° 58′ 11″ Ouest
Ouled Ziane	**3**	Latitude 33,2096° or 33° 12' 35" nord Longitude -7,197° or 7° 11' 49" ouest
Dhar Ben Allal	**4**	Latitude 34,84842° or 34° 50' 54" nord Longitude -6,21367° ou 6° 12' 49" ouest
Sidi Mohamed Lhmar	**5**	Latitude: 34.7167, Longitude: -6.26667 34° 43′ 0″ Nord, 6° 16′ 0″ Ouest
Ouled Mesbah Kbar	**6**	Latitude 34,98478° or 34° 59' 5" nord Longitude -6,17571° or 6° 10' 33" ouest
Ouled Mesbah Rouif	**7**	Latitude 34,98478° or 34° 59' 5" nord Longitude -6,17571° or 6° 10' 33" ouest
Lgnafda	**8**	Latitude 34,79089° or 34° 47' 27" nord

		Longitude -1,86661° or 1° 51' 60" ouest
Ezzaouia	**9**	Latitude 34.8853° or 34° 53' 7" north Longitude -5.81688° or 5° 49' 1" west
Laanabsa	**10**	Latitude 34,7808° or 34° 46' 51" nord Longitude -5,97478° or 5° 58' 29" ouest
Kariat Ben Aouda	**11**	Latitude 34,7698° or 34° 46' 11" nord Longitude -5,94808° or 5° 56' 53" ouest
Chouafaa	**12**	Latitude 34,76242° or 34° 45' 45" nord Longitude -6,04502° or 6° 2' 42" ouest

It should be noted that *B. truncatus* has been reported as the main intermediate host mollusk. *P. metidjensis* appears to be less common than *B. truncatus*. The former species has been reported in the Kénitra-Ouled Azzouz focus and in Sidi Allal Tazi. Other species have been reported in the plain, such as *Physa acuta* and *Lymnea peregra*.

Finally, all individuals subjected to the so-called cercarial emission test did not show infestation by *S. haematobium*. It therefore seems that, at the current state, the populations of *P. metidjensis* pose no human danger.

DISCUSSION

Among the 25 species of gastropod mollusks recorded across the entire Moroccan territory **(Ramdani *et al*., 1987; Saoud, 1995; Fadli, 2003; Walther *et al*., 2016)**, we identified 16 species in the Gharb plain. In the merja of Fouarates (a semi-permanent pond located near Kénitra), 10 species, which is more than half of the total number of freshwater gastropods in Morocco, were collected. The representativeness and species richness of gastropods are therefore very important in the plain.

Theoretically, one should expect a correlation between the geographical distribution of *P. metidjensis* foci (Fig. 2) and those of schistosomiasis foci (Fig. 3). However, the overlap of the Figs. (2, 3) shows that, in the Gharb plain, the presence of schistosomiasis foci is not necessarily accompanied by the presence of *P. metidjensis* foci. The reverse is also possible. The Gnafda area and the Kénitra area are two examples of this.

We did not collect *B. truncatus*, and only *P. metidjensis* was recorded. **Gaud (1958)**, **Doumenge *et al*. (1987)**, **Kharboua (1988-1994)**, **Laamrani (1994)**, **Zekhnini (1994)**, **Maqboul *et al*. (2014)**, and **Balahbib (2020)** report that the latter species is a potential intermediate host of *S. haematobium* in Morocco. In the different areas of the Gharb plain, the absence of bilharzia cases in the presence of vector foci may be due to the absence of *S. haematobium*, but the presence of bilharziasis foci in the absence of habitats for *P. metidjensis* led us to formulate several hypotheses:

- In the studied plain, some foci of schistosomiasis are allochthonous: it is possible that the schistosomiasis infestation of certain groups of inhabitants of the plain occurs in other regions of Morocco where schistosomiasis is endemic.

The geographical distribution of *P. metidjensis* is not stable over time. **Gaud (1952)**, **Benmansour (1970)**, **World Health Organization (1980)**, and **Sam-Ekobo (1984)** reported this type of instability in the populations of *Bulinus truncatus* **(Ernould, 1996)**. In this case, the climatic irregularity of the region could be the cause of this phenomenon. Indeed, **McCullough (1957)**, **Onabamiro (1972)**, **Sodeman (1979)**, **Saladin *et al*. (1980)**, **Coulibaly and Madsen (1990)**, **Perez Saez (2018)** and **Koperski (2022)** reported that the main factors controlling the dynamics of aquatic malacological populations, and consequently their spatio-temporal stability, are desiccation, temperature and precipitation. Furthermore, changes in ecological conditions due to climate change could lead to a shift in the geographic distribution of schistosomiasis **(Abdel-Wareth & Sayed, 2023)**.

- Finally, one last hypothesis that seems unlikely to us is the existence in the region of other types of vectors for *S. haematobium*: the gastropod mollusk that is currently common in all historically known sites as foci of bilharzia is the physid *Physa acuta*. Considering the potential of this species to be experimentally infested by *Schistosoma haematobium*, we can hypothesize that *P. acuta* is a third intermediate host of urinary bilharzia in Morocco.
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CONCLUSION

The population of gastropod mollusks in the Gharb plain is rich and diverse. The richness of the plain in various types and watercourses is certainly a cause of this wealth and diversity. However, throughout the recent years, *Bulinus truncatus*, the main vector of urogenital schistosomiasis in Morocco, has not been present in the malacological population of the plain. This absence could be confirmed by a lack of human cases infested by *Schistosoma haematobium*, the flatworm responsible for human urogenital schistosomiasis. However, this absence of schistosomiasis in the studied plain raises several epidemiological issues, namely:

- Although *Planorbis metidjensis* existed until 1998 among the collected population, no cases of indigenous human schistosomiasis have been detected in the plain since 1972.
- Theoretically, we should expect to find a correlation between the geographical distribution of *P. metidjensis* foci and those of bilharzia foci.
- Historically, several schistosomiasis foci and the presence of *B. truncatus* among the fauna of the plain have been reported.
- Since 2000, after the disappearance of *B. truncatus, P. metidjensi* is no longer part

of the fauna of the plain. The disappearance of these two species in the Gharb plain remains an ecological and epidemiological enigma, especially since their disappearance has not been followed by any chemical treatment of the environment.

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