

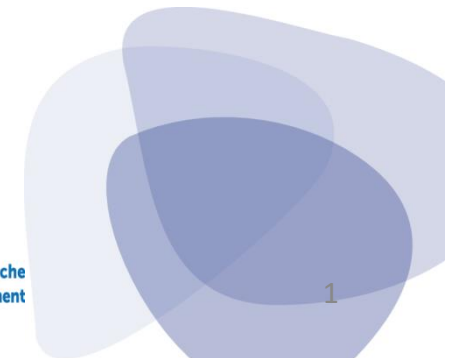
Ecology of schistosome species interactions and involvement of rodents in the transmission

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MCF/HDR / en délégation IRD au Bénin

Niamey le 29/11/2022



Lineage

Eukaryota

Opisthokonta

Animalia

Eumetazoa

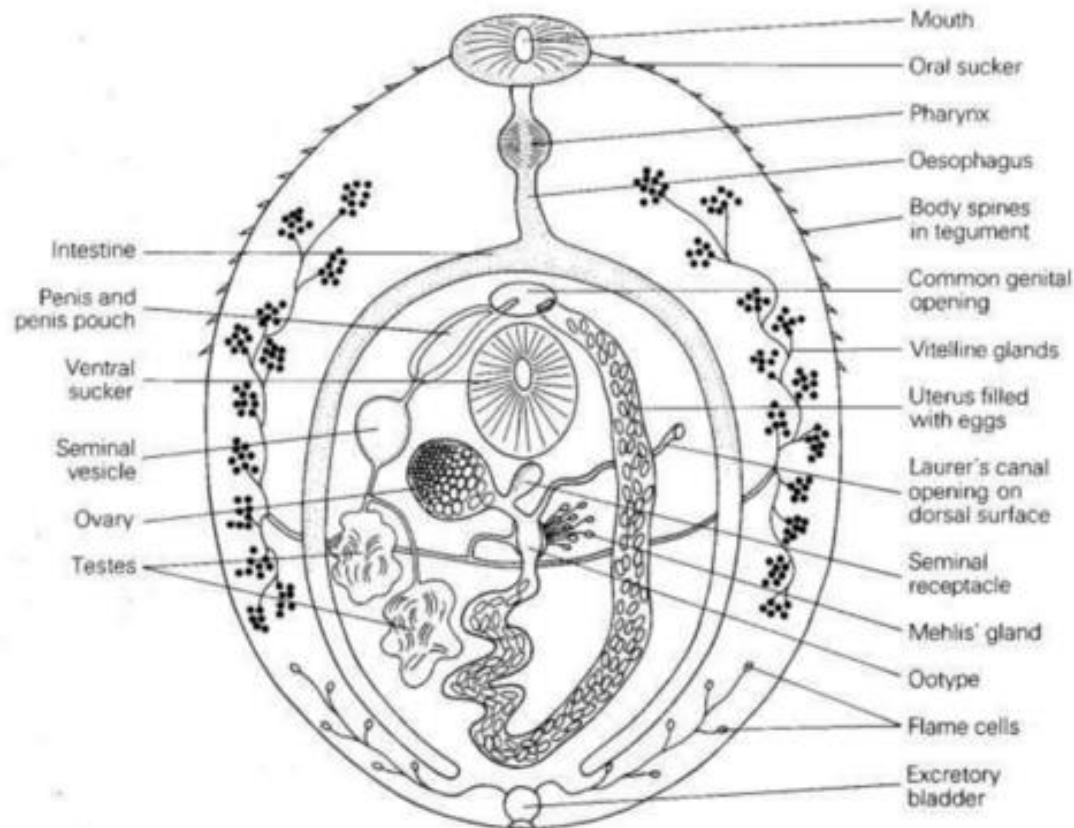
Bilateria

Protostomia

- Phylum: Plathelminthes
- Class: Trematoda (blowhole = évent)
- Subclass: Digenea
- Order: Diplostomida (two suckers)
- Superfamily: Schistosomatoidea
- Family: Schistosomatidae
- Genus: *Schistosoma*

Most platyhelminths are **hermaphroditic**, with each individual possessing both male and female reproductive organs ...

... except schistosomes that are **gonochoric**.



<i>Schistosoma species</i>	Definitive host
<i>S. japonicum</i>	Human and other Mammals in China, Philippines, Japon, ...
<i>S. mekongi</i>	Human along the Mekong (Laos and Cambodia)
<i>S. malayensis</i>	Human and murids in Malaysia
<i>S. sinensium</i>	<i>Rattus rattus</i> et <i>Crocidura attenuata</i> in China and Thaïland
<i>S. ovuncatum</i>	<i>Mus musculus</i> (Labo); Northwest of Thaïland

<i>S. hippopotami</i>	Hippopotamus in Ouganda
<i>S. edwardiense</i>	

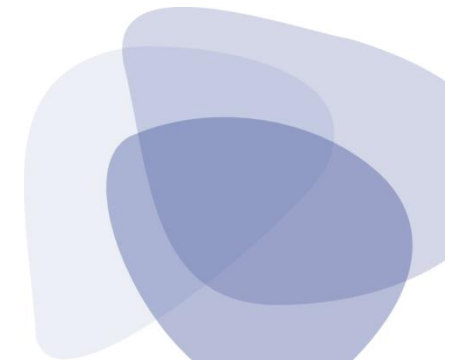
<i>S. incognitum</i>	Mammals including murids (Thaïland, Indonesia...)
<i>S. turkestanicum</i>	Ruminants in Asia and Hungary (red deer = Cerf élaphe)

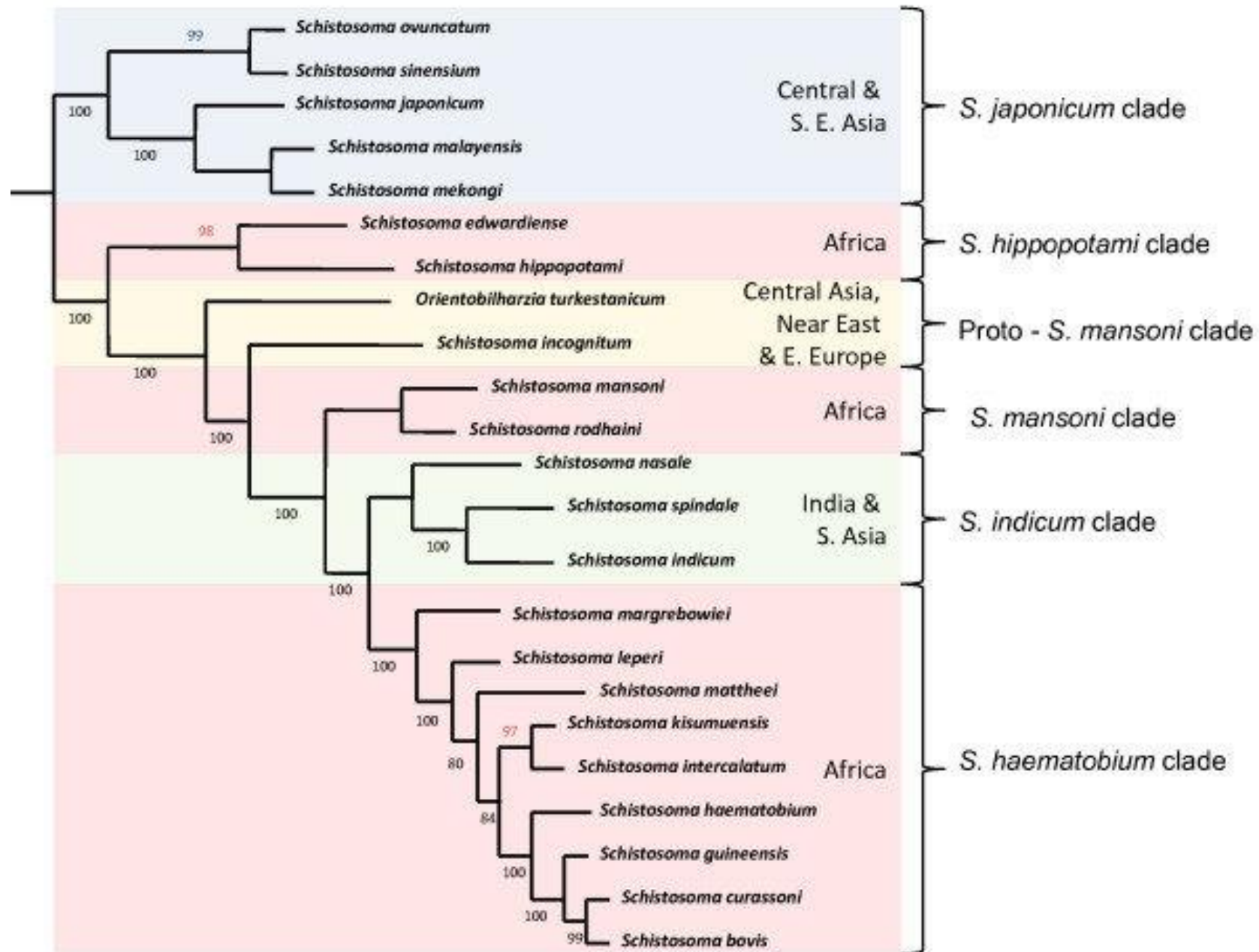
<i>S. mansoni</i>	Human in Africa, Middle East and tropical America
<i>S. rodhaini</i>	Murids in Africa

<i>S. indicum</i>	Cattle and other Mammals in India
<i>S. spindale</i>	Cattle in India and neighboring countries
<i>S. nasale</i>	

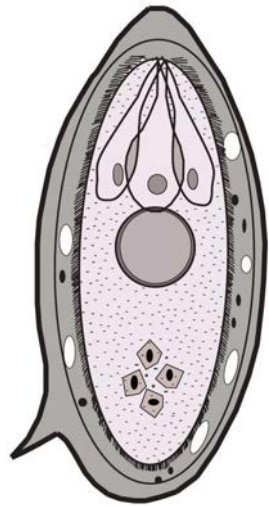
Terminal spine egg group

<i>Schistosoma</i> species	Definitive host
<i>S. bovis</i>	Domestic and wild bovids
<i>S. curassoni</i>	Sheep and goats in West Africa
<i>S. leiperi</i>	Antelopes and domestic cattle in Africa
<i>S. margrebowiei</i>	
<i>S. kisumuensis</i>	Murids in Kenya
<i>S. mattheei</i>	Human, antelopes and domestic cattle in Africa
<i>S. haematobium</i>	Human in Africa, Middle East and Corsica (France)
<i>S. guineensis</i>	Human in the Gulf of Guinea
<i>S. intercalatum</i>	Human in Central Africa





Phylogeny estimated with a Bayesian analysis of combined partial *l*srDNA (Lipolysis-stimulated lipoprotein receptor), complete *ssr*DNA (simple sequence repeats or μ sat) and partial *cox1* (Lawton et al. 2011)



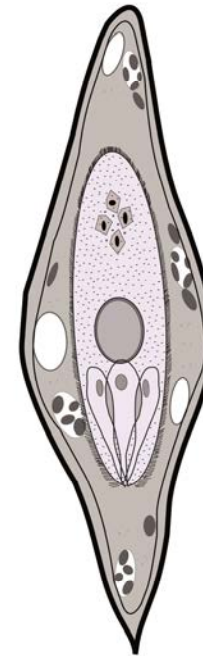
S. mansoni



S. rodhaini



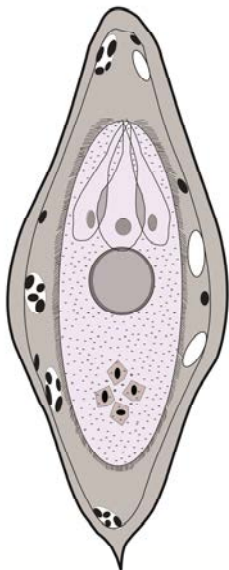
S. raematobium



S. bovis



S. curassoni



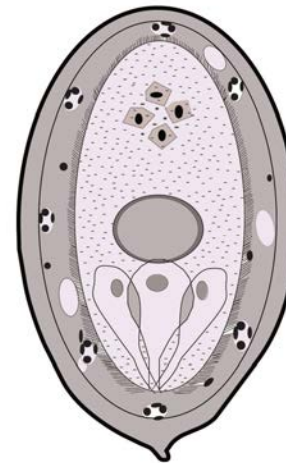
S. intercalatum
S. guineensis



S. matthei

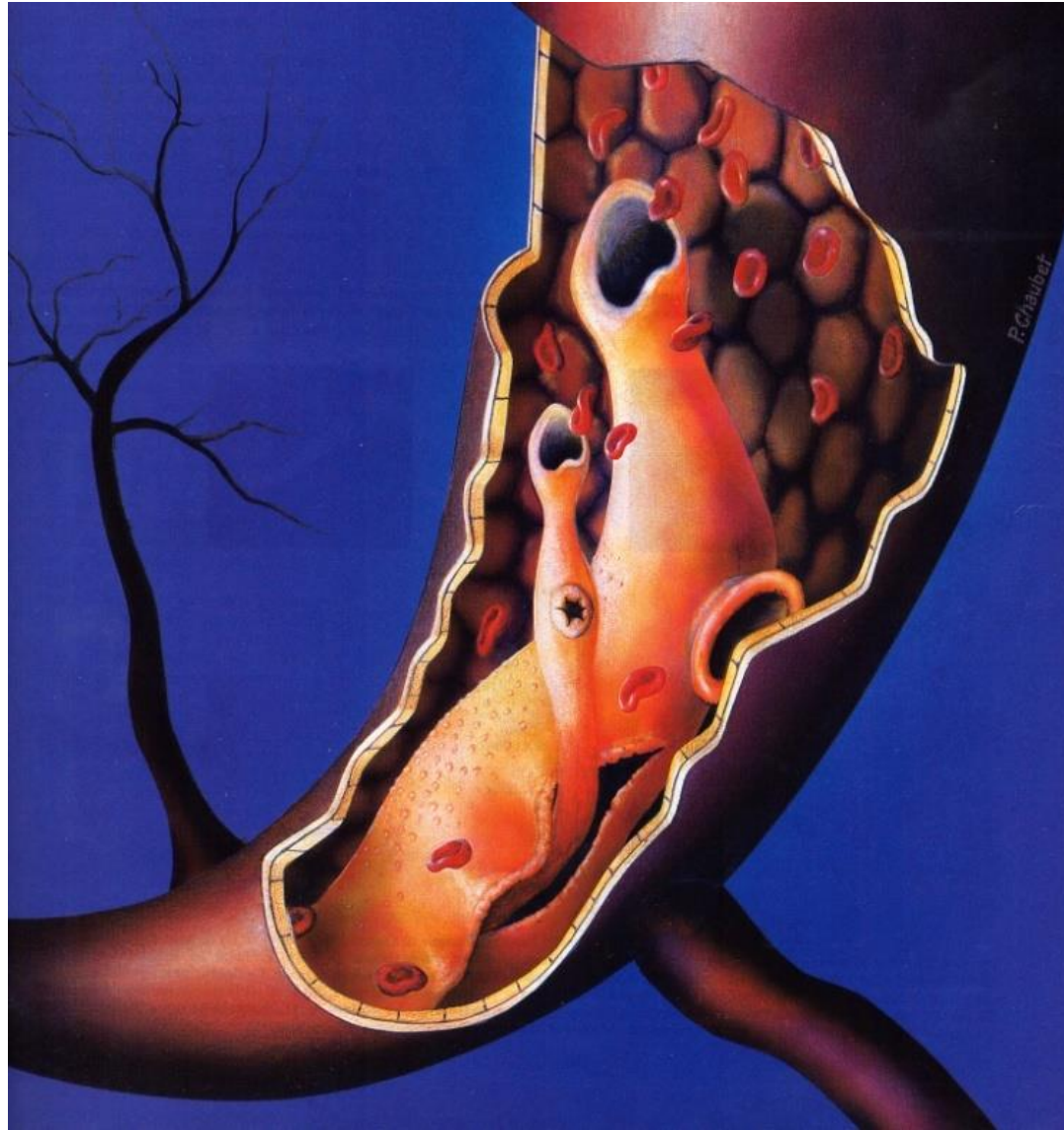


S. leiperi



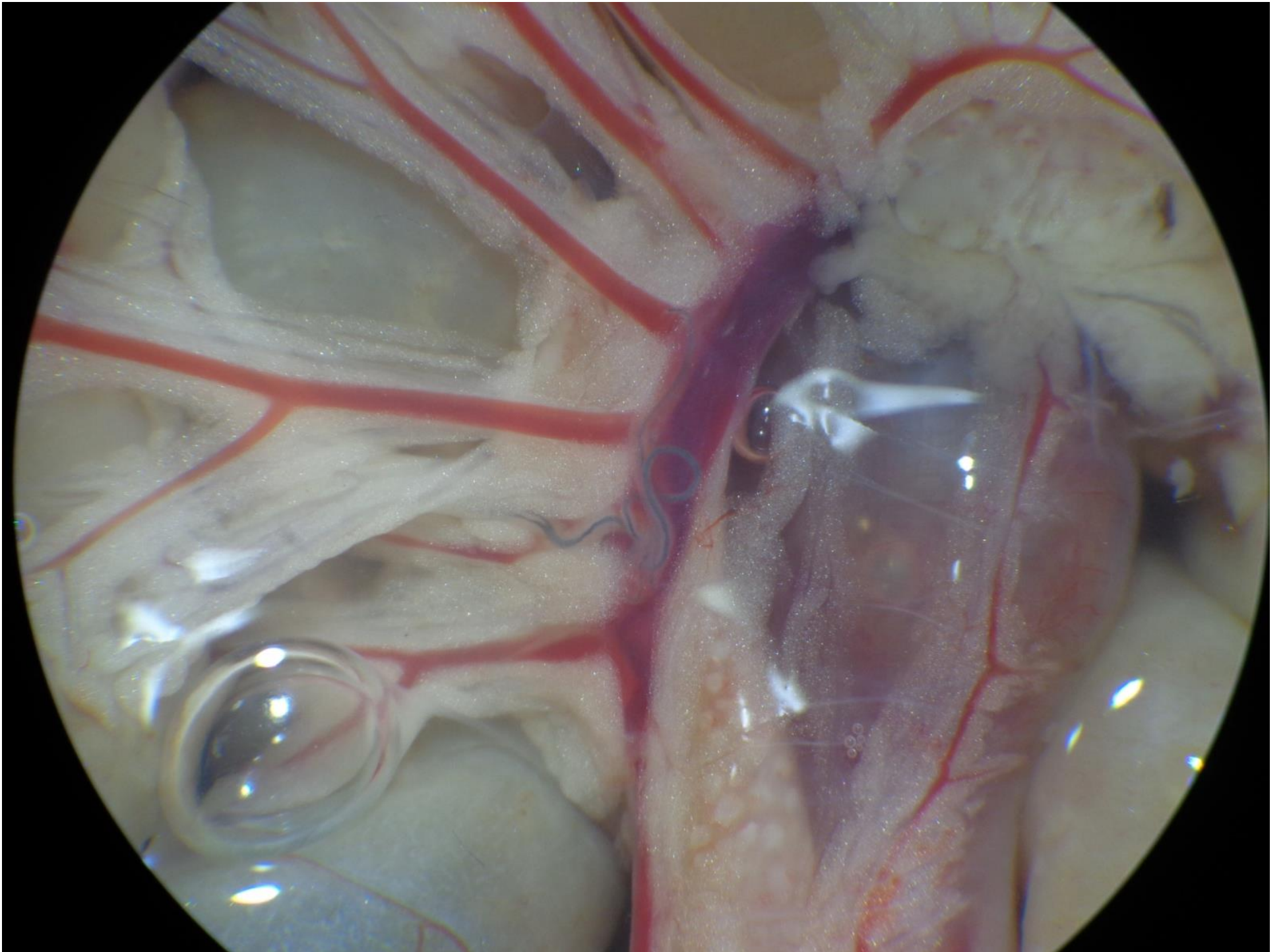
S. margrebowiei

By Gabriel Mouahid
Not published



A pair of schistosomes in the mesenteric venous system of homoeothermic vertebrates

Painted by students from the School of Fine Arts in Perpignan

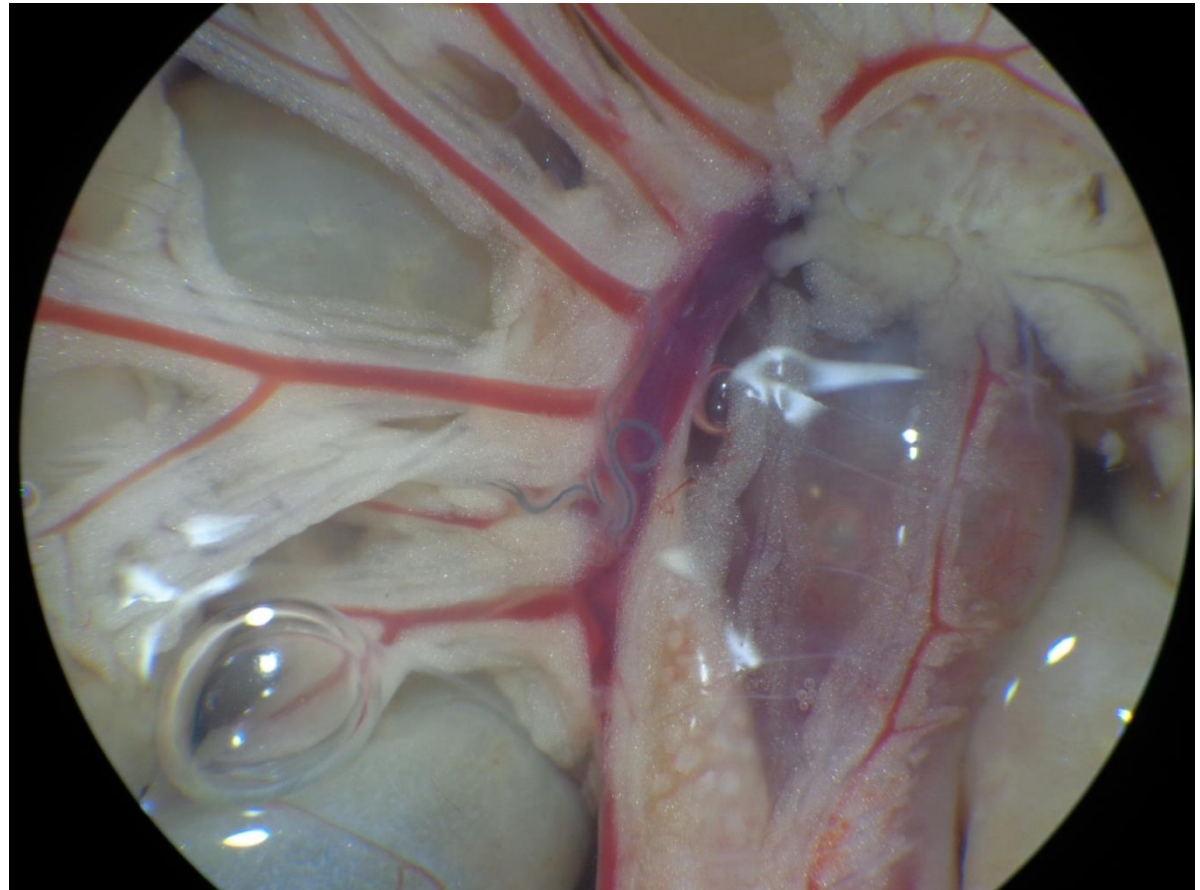


Schistosomatidae share the ability to live in the circulatory system of vertebrates with two groups of trematodes:

1. **Spirorchiidae** of turtles and
2. **Sanguinicolidae** of fish.

Parasites that live in intimate contact with the immune system of their hosts require **specialized adaptations** to survive in such exposed environments.

(Brant & Loker, 2005 *PLoS Pathogens*)

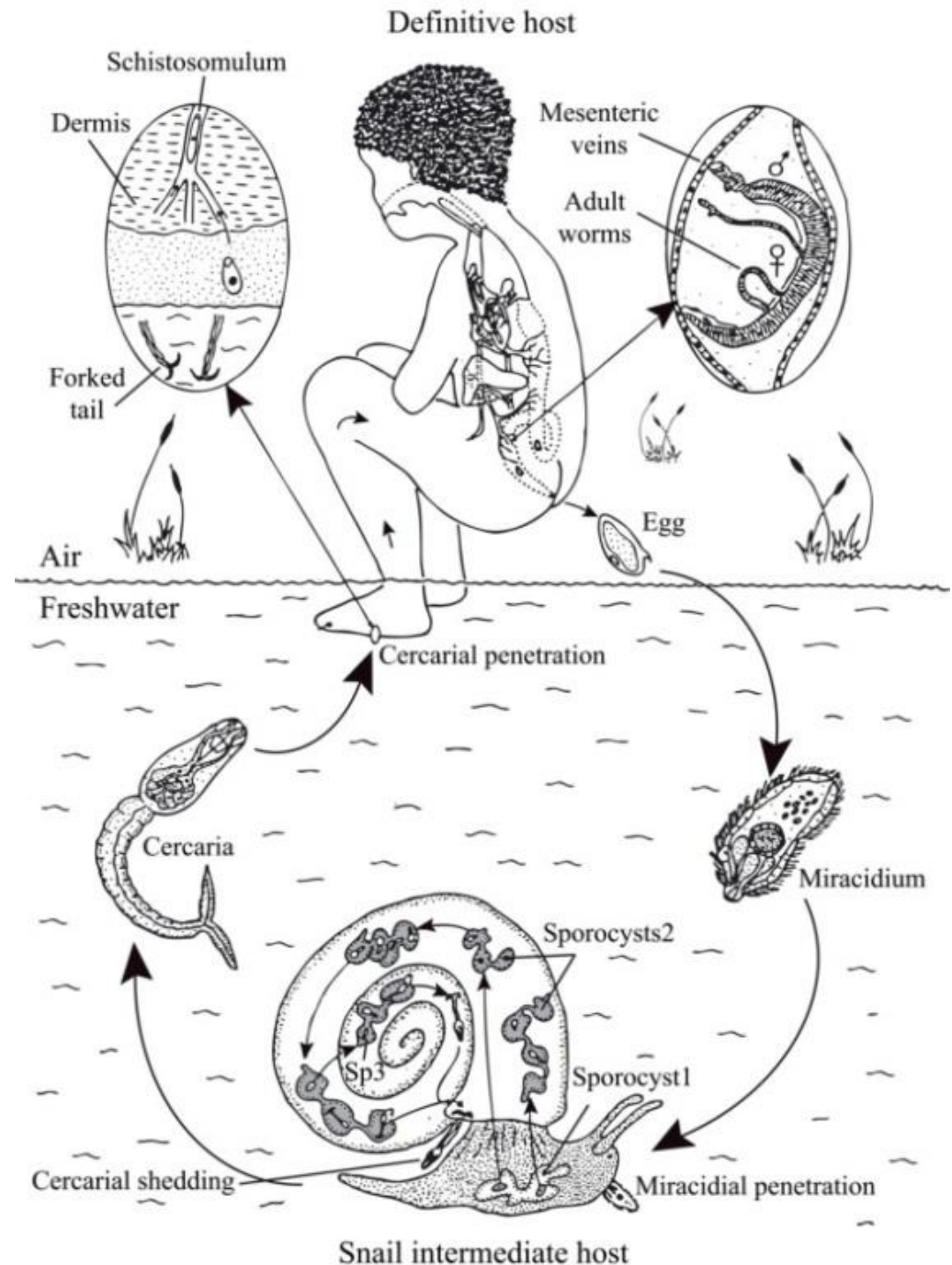


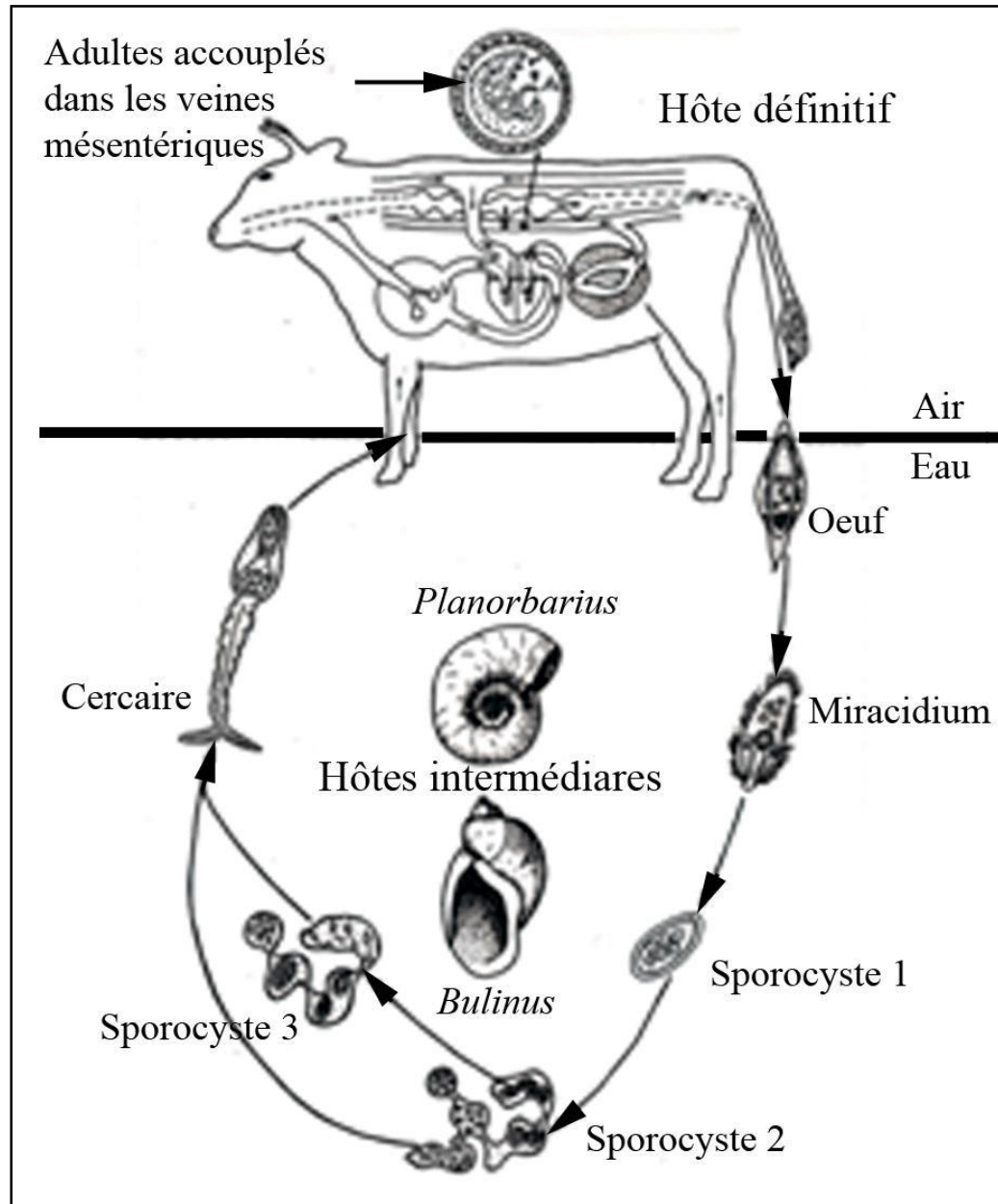
Complex life cycle

Schistosoma mansoni, causative agent of human intestinal Bilharziasis

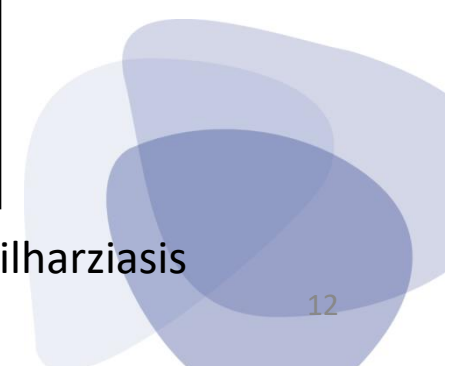
By Dr H  l  ne Mon  

Schistosomes alternate sexually and asexually reproducing generations in their life cycle.





Life cycle of *Schistosoma bovis*, causative agent of intestinal Bilharziasis
 By G. Mouahid



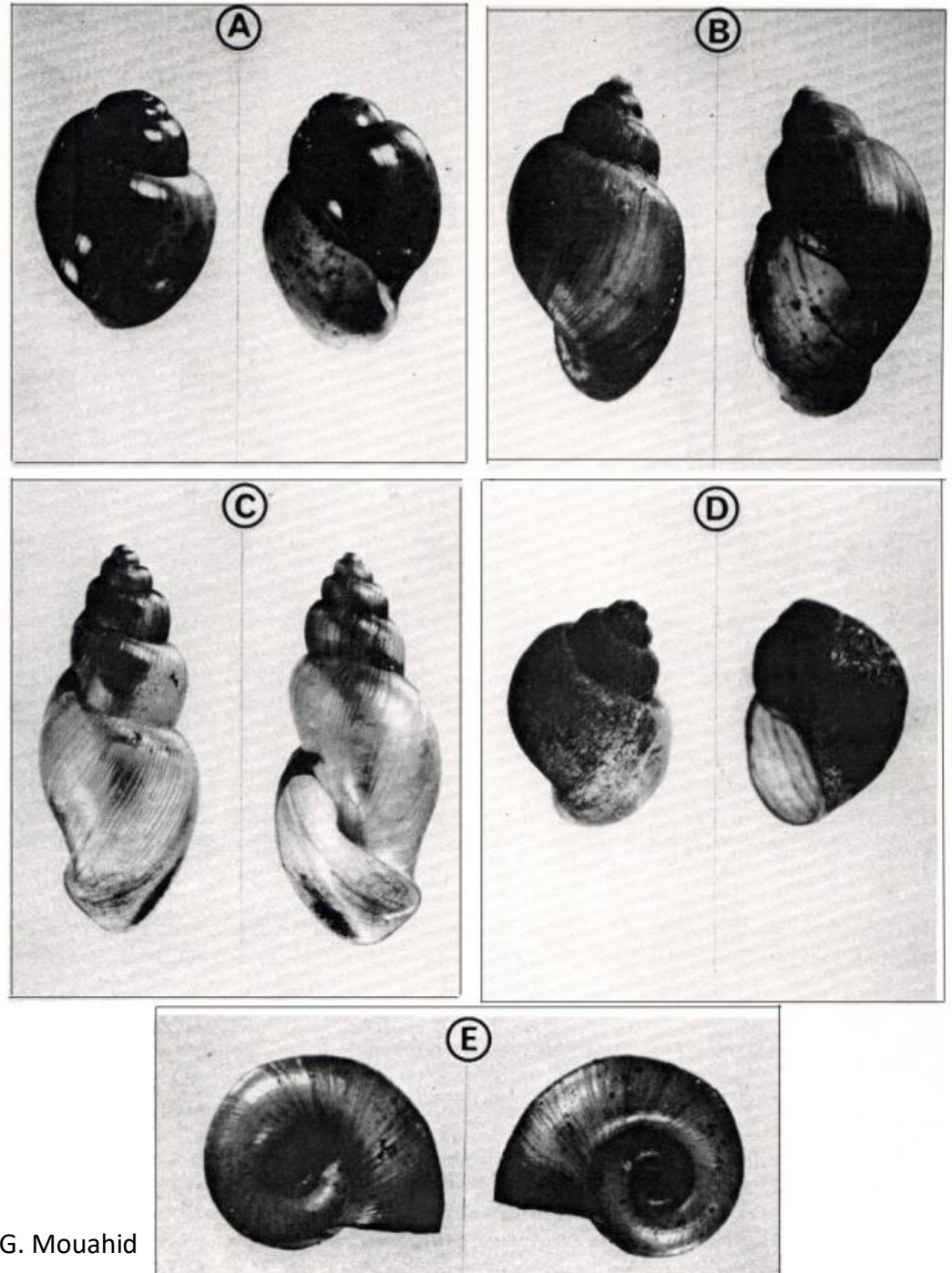
Freshwater snails implicated as intermediate hosts in the transmission of the terminal spine egg group of *Schistosoma* genus

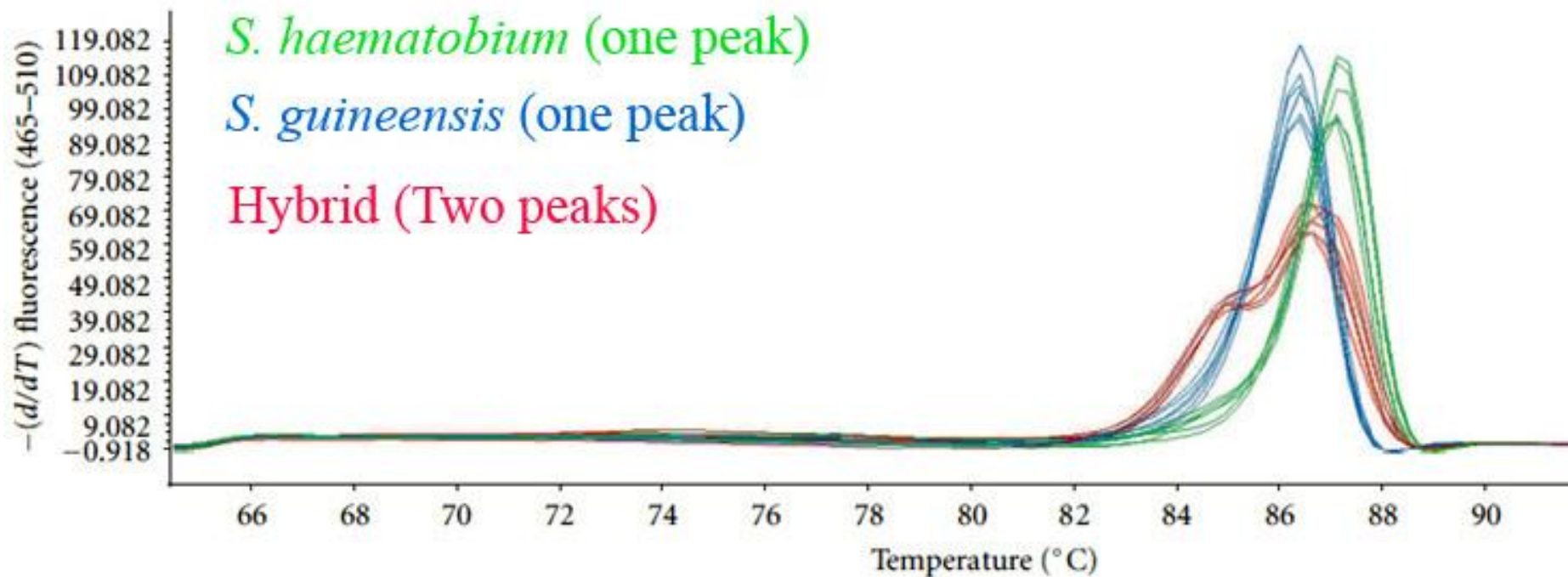
Bulinus genus:

- " *africanus* group (A)
- " *truncatus* / *tropicus* group (B)
- " *forskalii* group (C)
- " *reticulatus* group (D)

Planorbarius genus:

- " *Planorbarius metidjensis* (E)



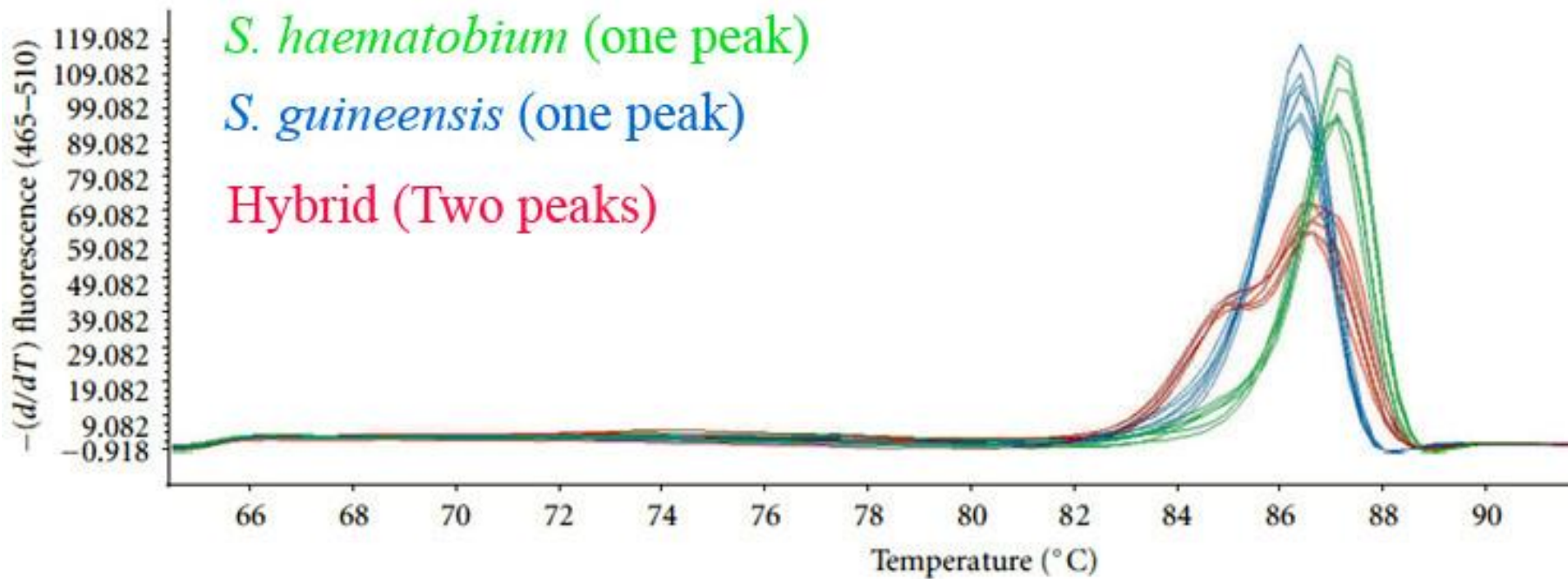


High resolution melting analysis (HRM). Melting peaks of 6 randomly selected individuals from the *Schistosoma guineensis* (blue), *S. haematobium* (green) and hybrid (red) patterns.

HRM is a PCR-based method for detecting DNA sequence variation, which enables detection of both homozygous and heterozygous sequences.

HRM analysis serves to distinguish between several PCR amplicons with subtle changes in sequence, down to the single nucleotide level.

Moné H., Minguez S., Ibikounlé M., Allienne J.-F., Massougbojji A. & Mouahid G. 2012. Natural interactions between *S. haematobium* and *S. guineensis* in the Republic of Benin. *The Scientific World Journal*. ID 793420. doi:10.1100/2012/793420.



<i>S. haematobium</i>	<u>G C A T A T C A A C</u>	<u>G C G G G T T G C T</u>	G G T C G A A G G C	T C C G T C C T A A	T A G T C C G G C C
<i>S. guineensis</i> A
			★		
<i>S. haematobium</i>	A C A G C C T A G T	C C G G T C T A G A	T G A C T T G A T C	G A G A T G C T G C	G G T G G G T T G T
<i>S. guineensis</i> T
			★		
<i>S. haematobium</i>	G C T C G A G T C G	T G G C T T A A T G	A C A T T A T A C G	C G C T C G G G A A	G A A T C G C A C C
<i>S. guineensis</i> A
			★		
<i>S. haematobium</i>	T A T C G T A C G C	T A C G T T G G T C	A C T T G A T C T T	G T C T C T A T G G	T T C G G T C T A C
<i>S. guineensis</i>
<i>S. haematobium</i>	<u>G G T T T G T</u>				
<i>S. guineensis</i>				

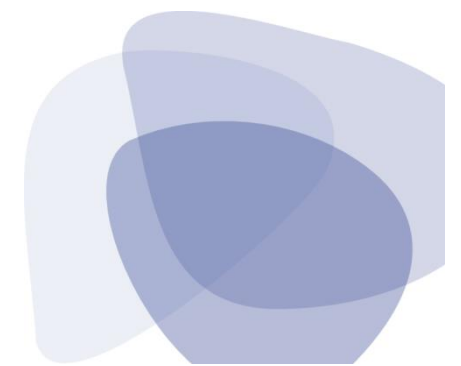
ITS2 fragment sequences showing the three single-nucleotide polymorphisms between *S. haematobium* and *S. guineensis*. The priming sites are underlined.



Maximum likelihood tree topology of **Cox1 mtDNA** (955bp) showing that *S. haematobium* from Corsica and also from Benin-Sô-Tchanhoué belongs to the *S. bovis* clade.

The scale shows the number of nucleotide substitutions per site.

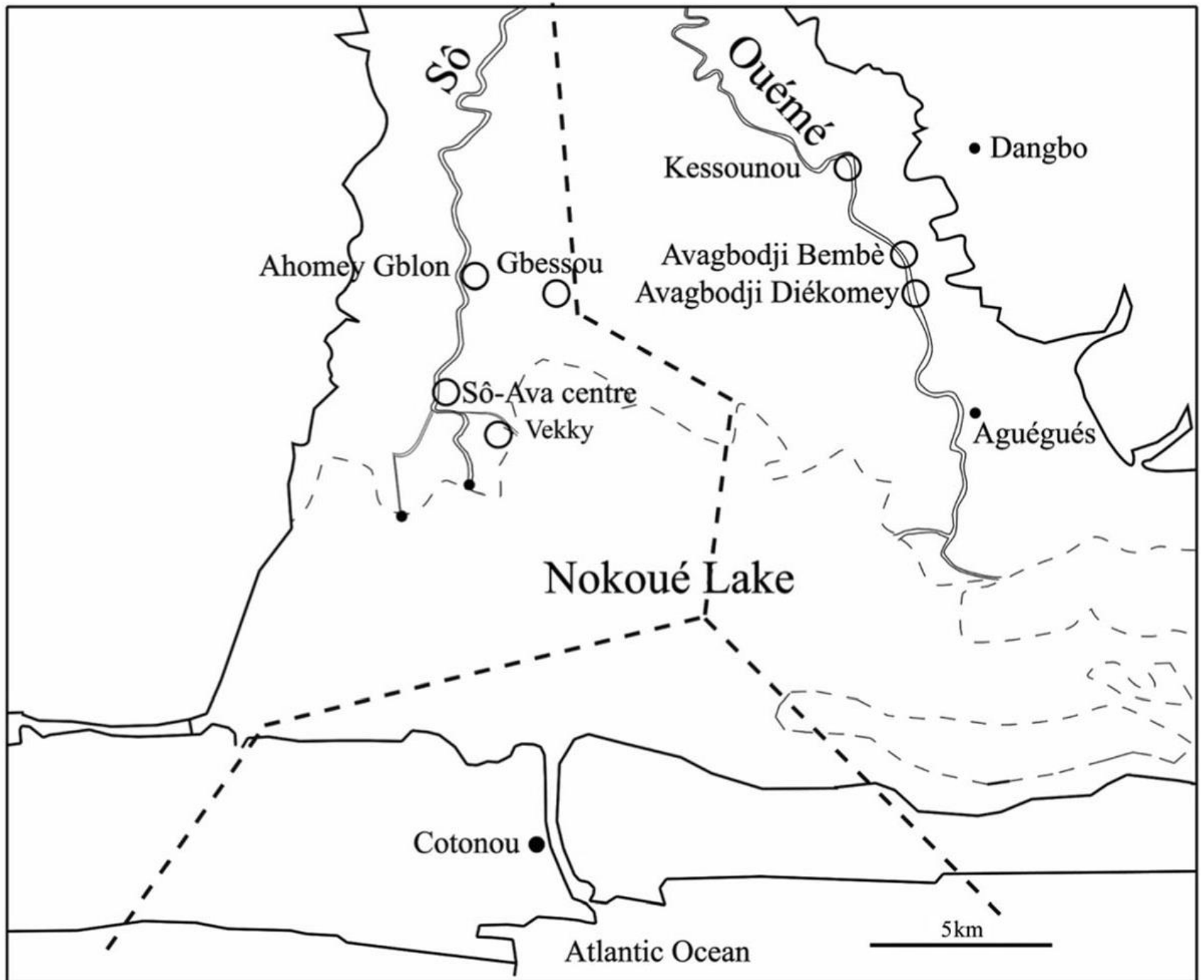




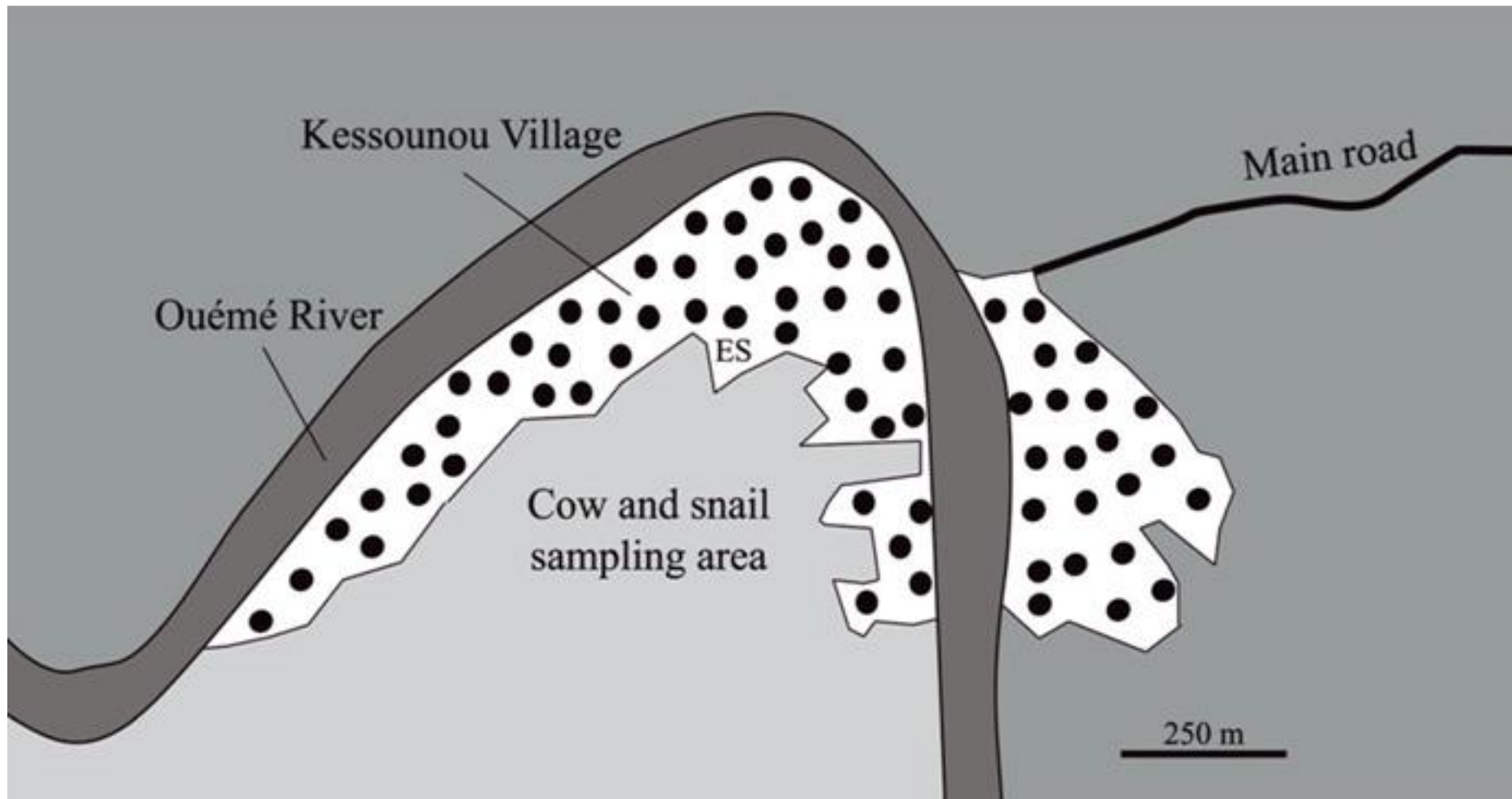


Study area





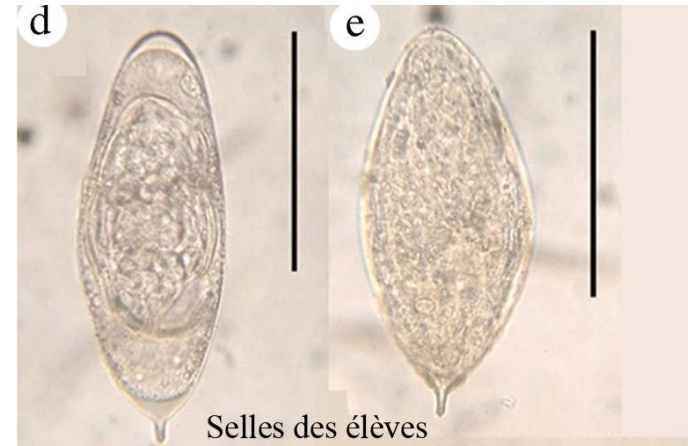
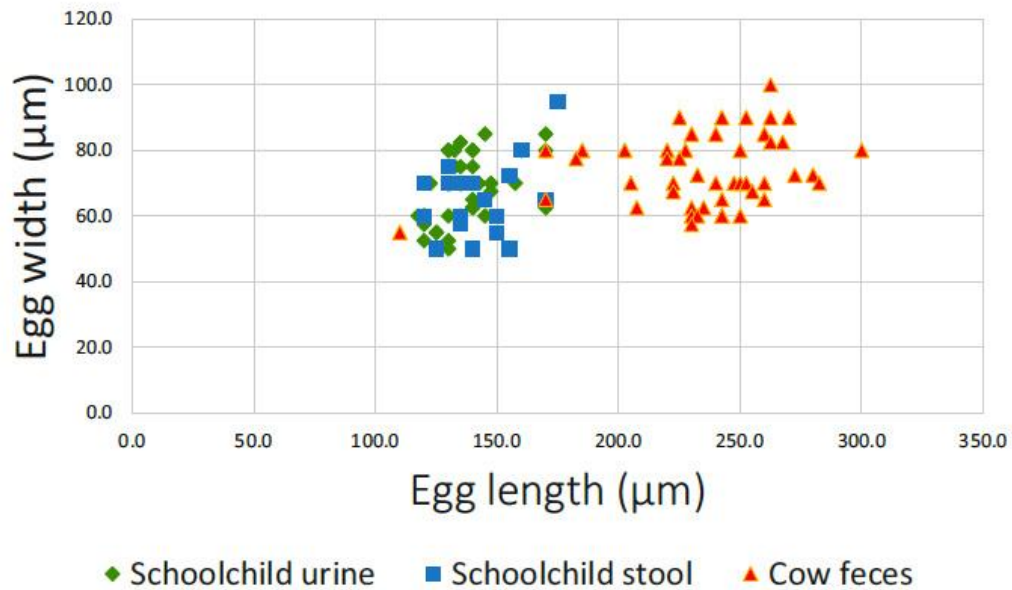
Kessounou: an area where rodents, humans and their livestock share the same habitat



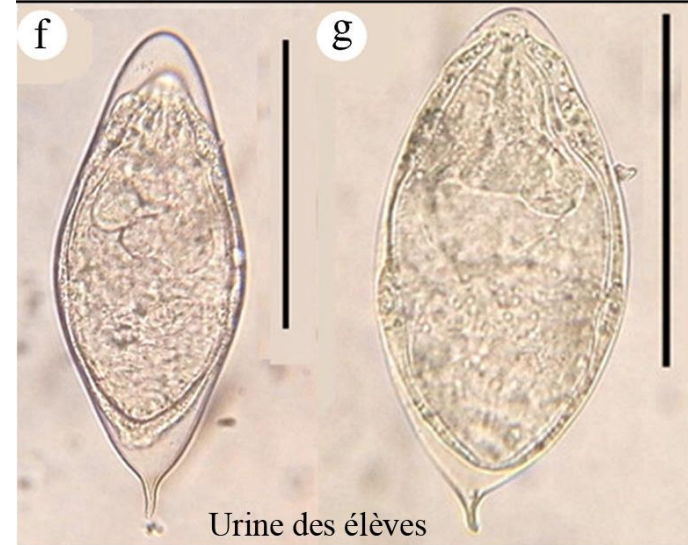
ES: Elementary School
Black dots represent the houses



Schistosoma eggs collected in cow feces
The scale represents 100 μm

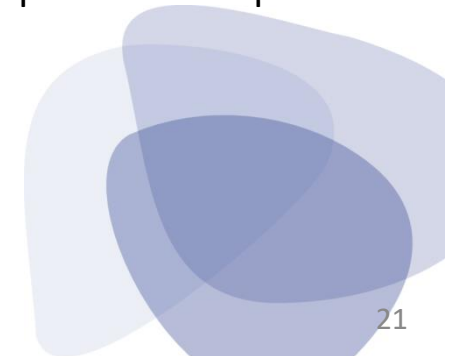


Selles des élèves



Urine des élèves

The scale represents 100 μm



Cox1

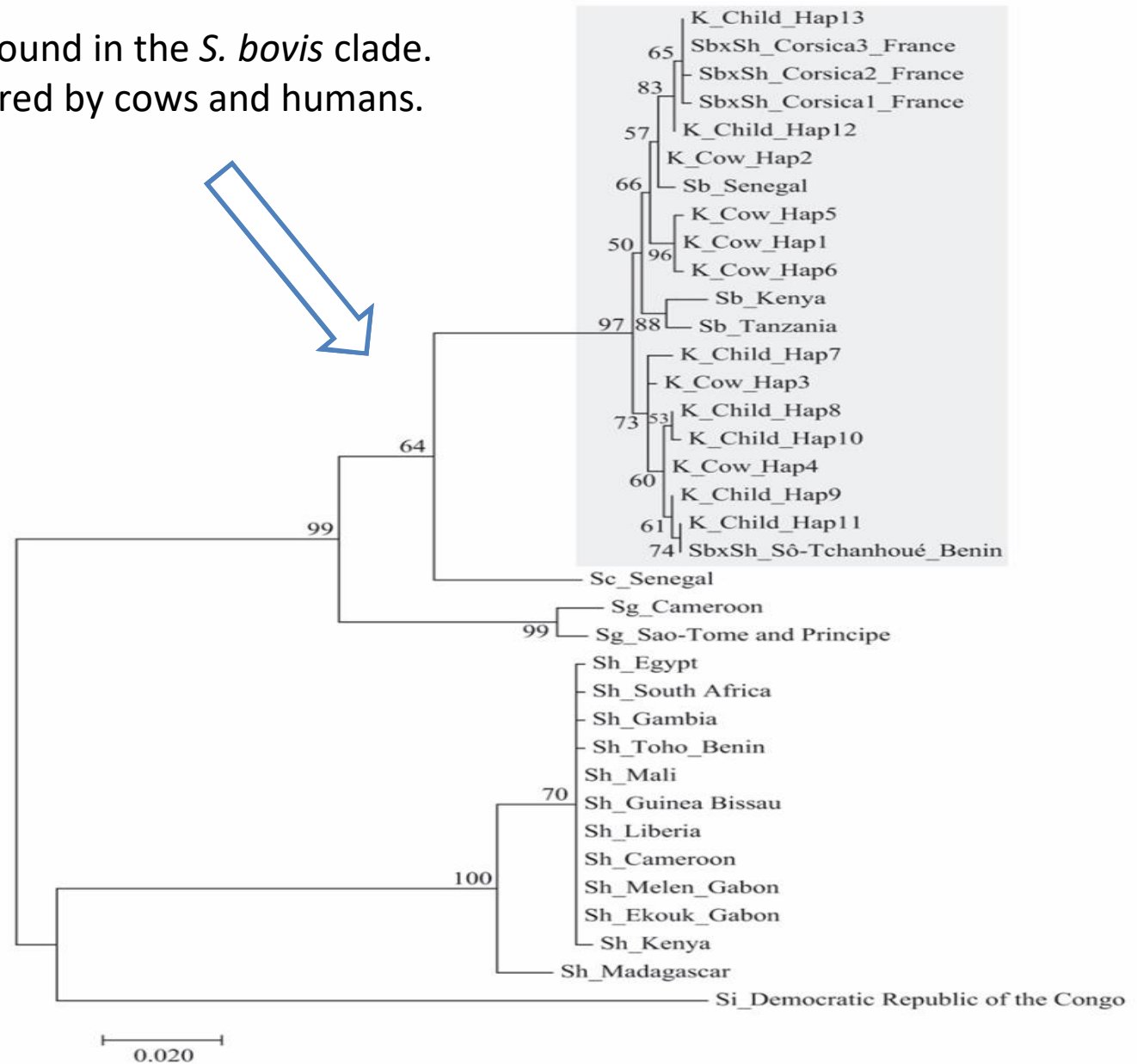
1. All Cox1 sequences are found in the *S. bovis* clade.
2. Cox1 haplotypes are shared by cows and humans.

ITS

- " *S. haematobium* ITS sequences are found only in humans
- " *S. bovis* ITS sequences are found only in cows.
- " "Hybrid" ITS sequences are shared by cows and humans.

First cases

- " of *S. bovis* in Benin
- " of presence of "hybrids" shared by cows and humans in the same village.

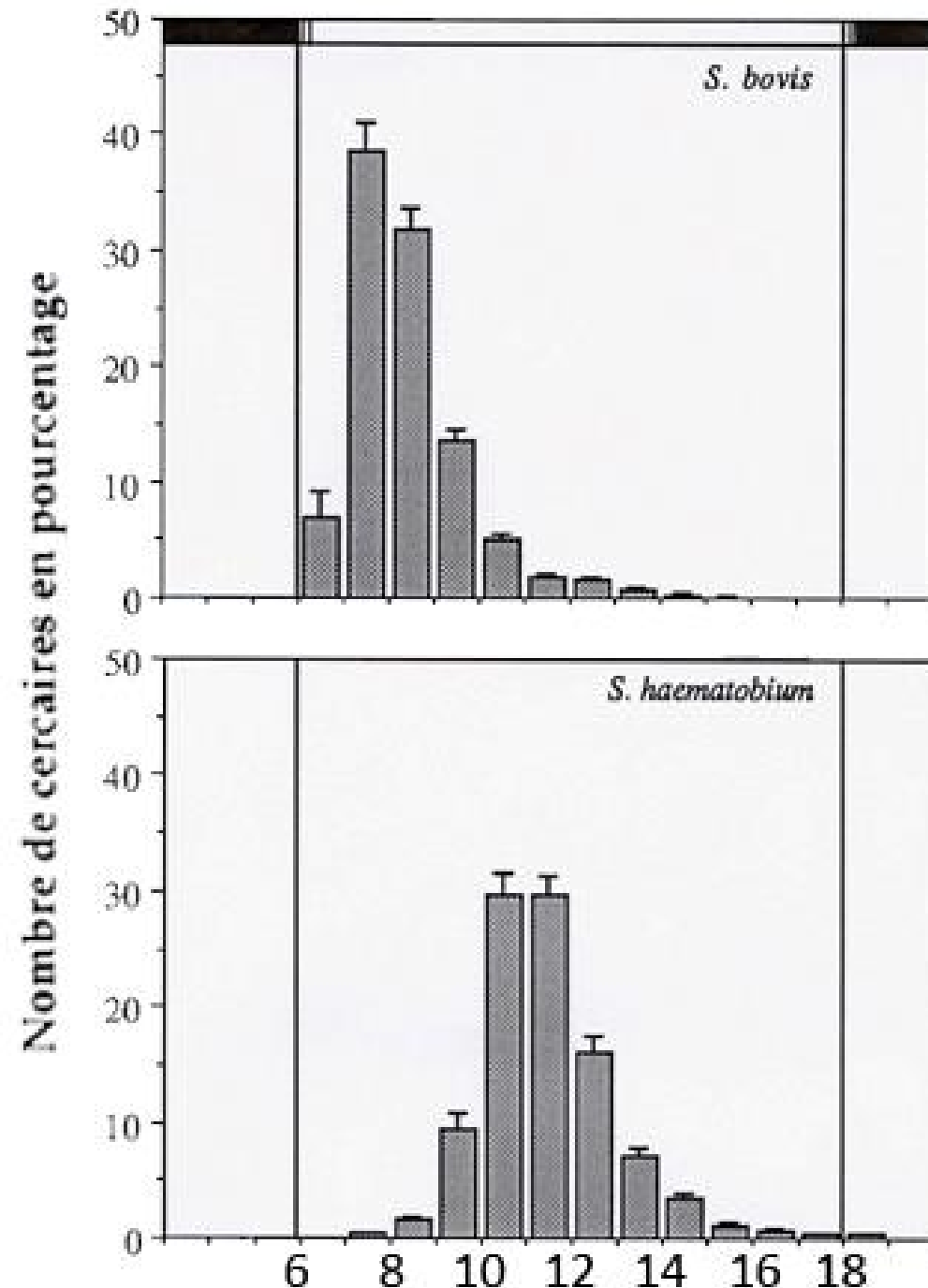


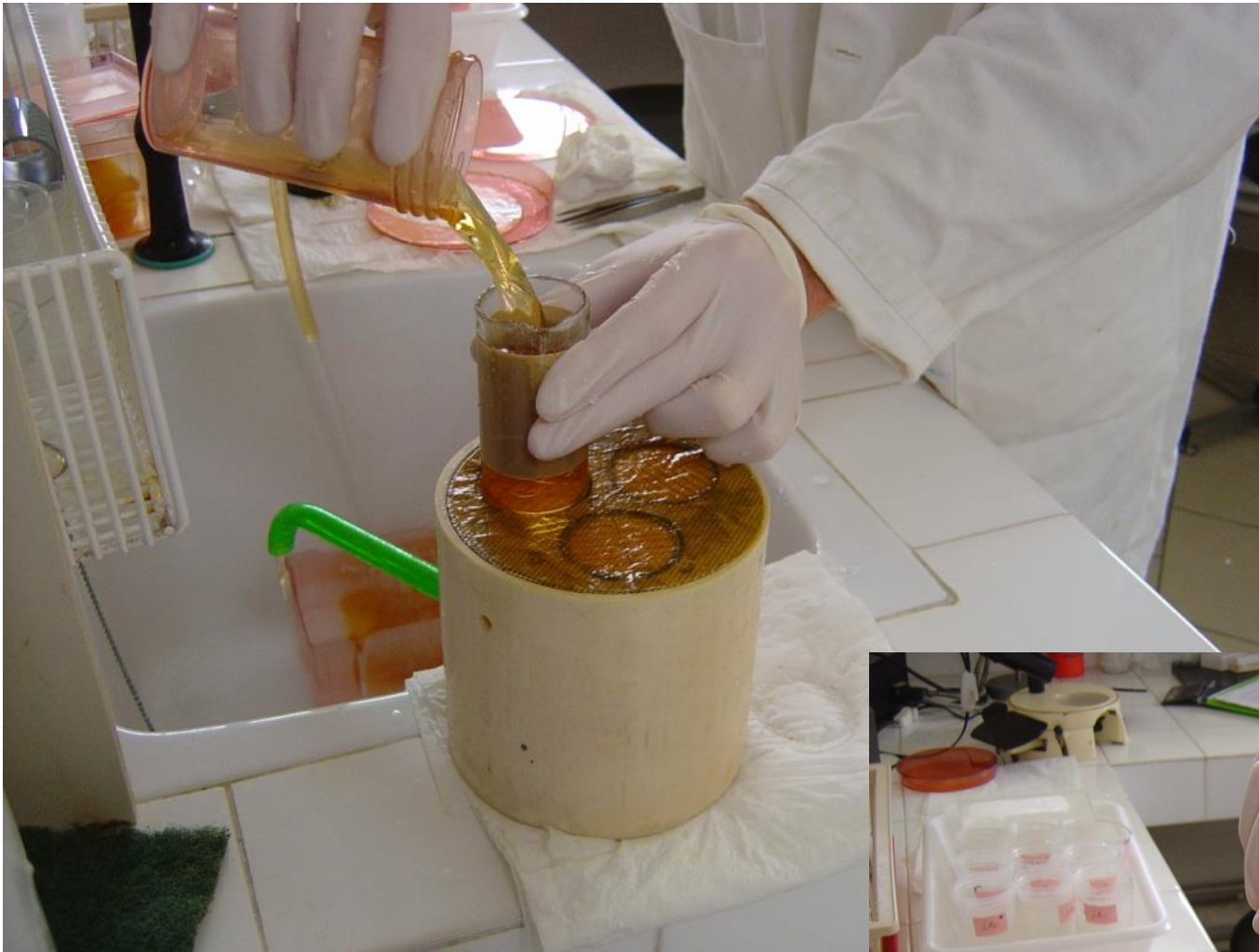
Savassi B., **Mouahid G.**, Lasica C., Mahaman SDK., Garcia A., Courtin D., Allienne JF., **Ibikounlé M.** & **Moné H.** 2020. *Parasitology Research*.

Since the two species are in interaction we **should expect** a cercarial emission pattern that takes into account crossbreeding.

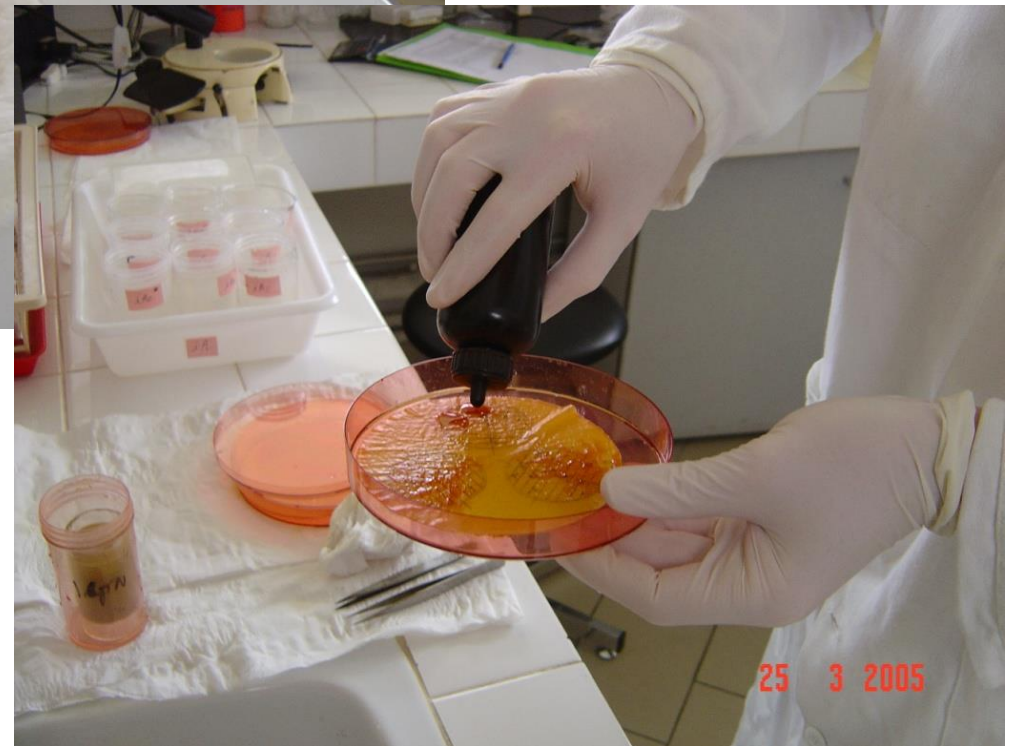
Cercarial emission pattern of *S. bovis* from Spain (above) and *S. haematobium* from Morocco (below).

Mouahid et al. 1991.
Journal of Helminthology



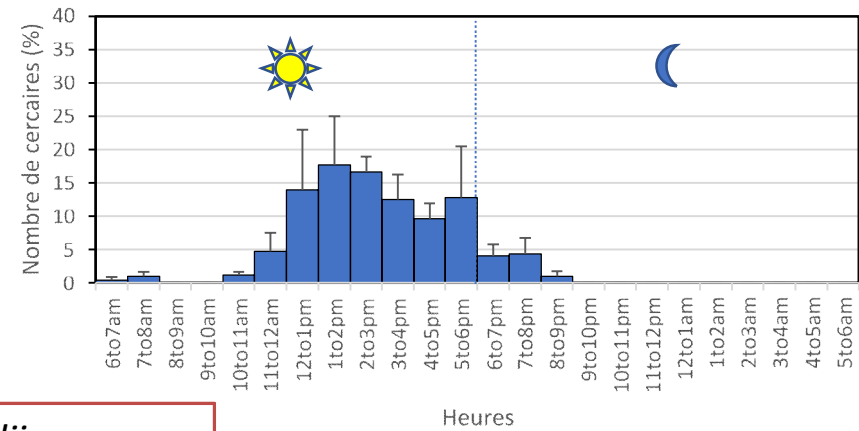
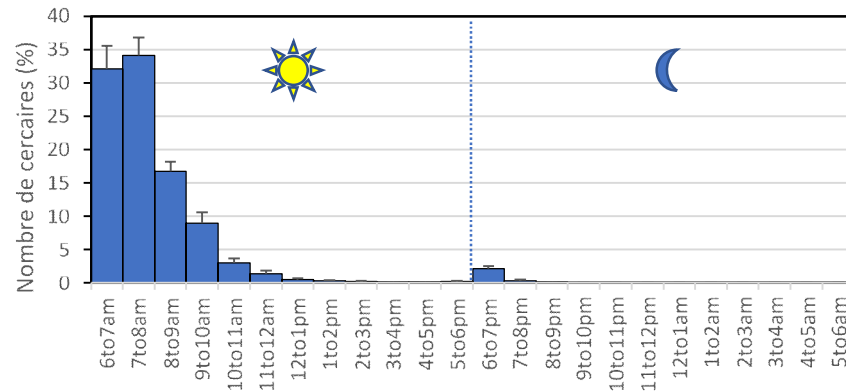


Cercarial emission patterns from infected snails (*Bulinus forskalii* and *Bulinus globosus*), each exposed to 3 miracidia of *Schistosoma* from cow feces.





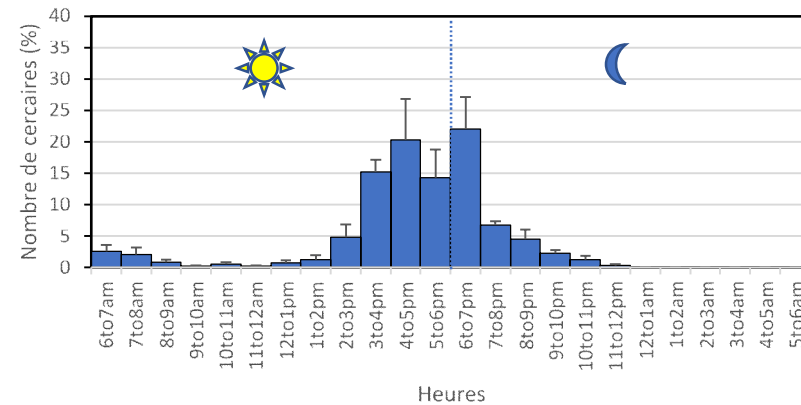
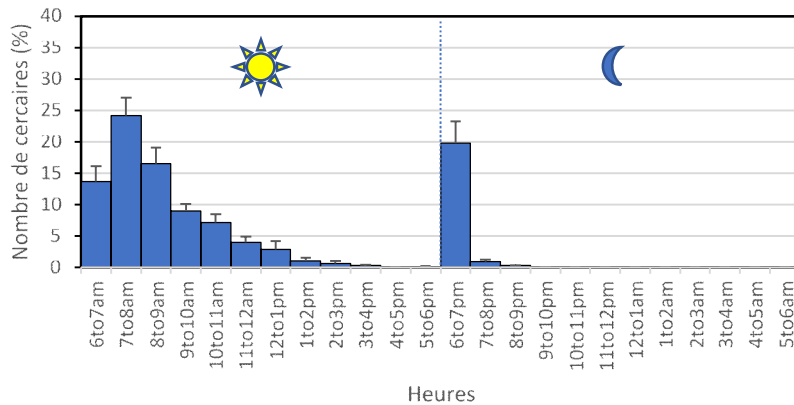
Observation and counting of cercariae with a dissecting microscope



a- Typical early diurnal pattern of *S. bovis* on *B. forskalii*

b- Typical late diurnal pattern of *S. haematobium* on *B. globosus*

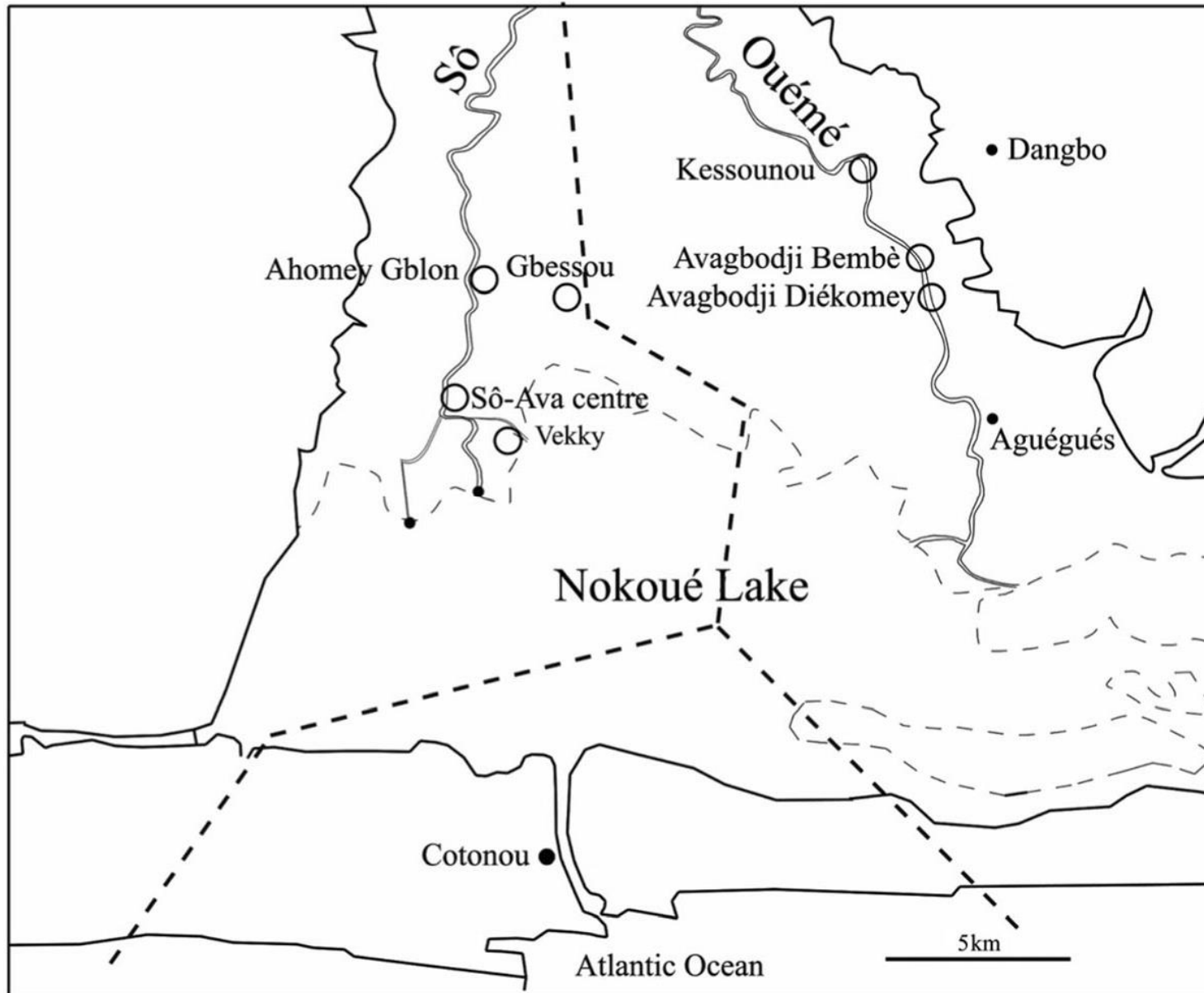
Diversity of cercarial emission patterns obtained in cattle



c- Early diurnal and nocturnal pattern on *B. forskalii*

d- Late diurnal and nocturnal pattern on *B. globosus*

How to explain the nocturnal emission?





Gabriel Mouahid

Voyage en pirogue avec tout le matériel pour 3 jours



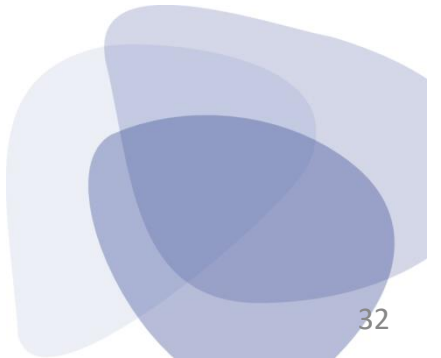
Pose des pièges dans les foyers







The village of Gubessou is part of the district of Houédo-Aguékon





Laboratoire de Recherche en Biologie Appliquée, Unité de Recherche sur les Invasions Biologiques; Ecole d'Abomey-Calavi, Université d'Abomey-Calavi, Benin



a : *Rattus norvegicus*, b : *Rattus rattus* ; c : *Mastomys natalensis*; d : *Crocidura olivieri*

- A total of 61 captures (including 50 perfused rodents) out of 499 traps set (12.2%).
- 6 rodents positive for schistosomes, i.e. 12% prevalence of infestation.



Laboratoire de Recherche en Biologie Appliquée, Unité de Recherche sur les Invasions Biologiques; Ecole d'Abomey-Calavi, Université d'Abomey-Calavi, Benin

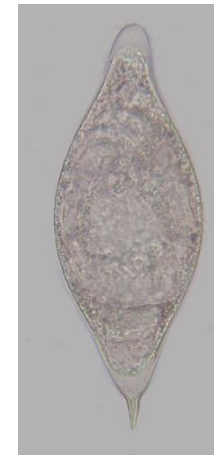


The recovery of schistosomes is done by perfusion. The method used was developed by Duwall & Dewitt in 1967. The infusion solution is composed of 7.5 g/l sodium citrate and 8.5 g/l sodium chloride.

Schistosome egg
morphotypes from
Mastomys natalensis



a



b

Schistosome egg
morphotypes from
Rattus rattus



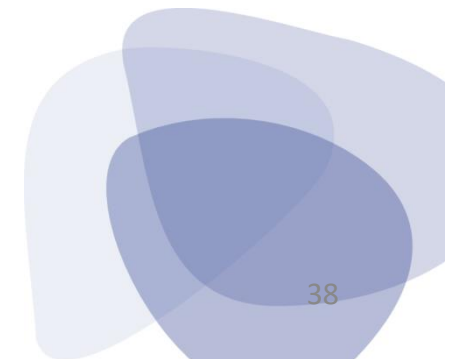
c

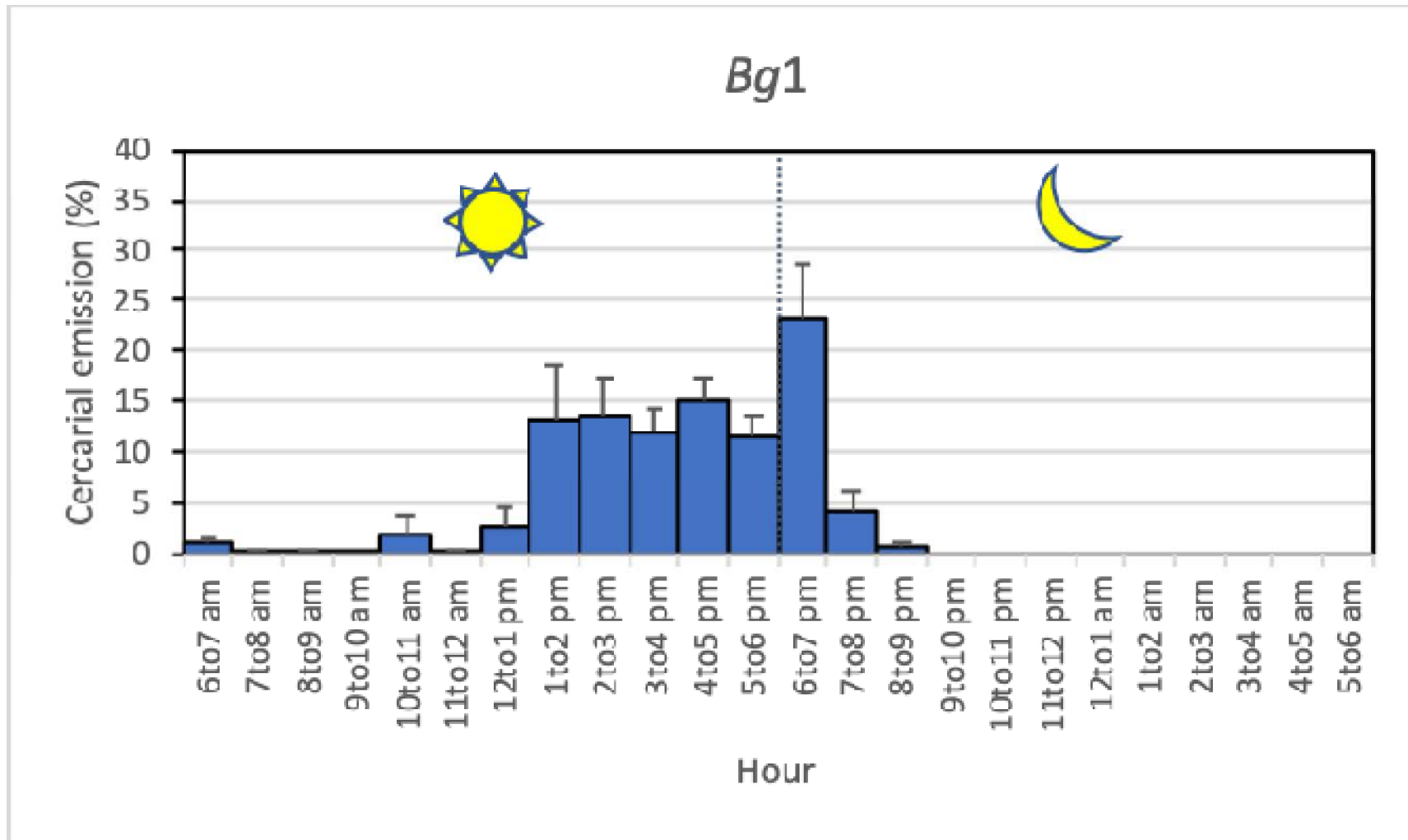


d

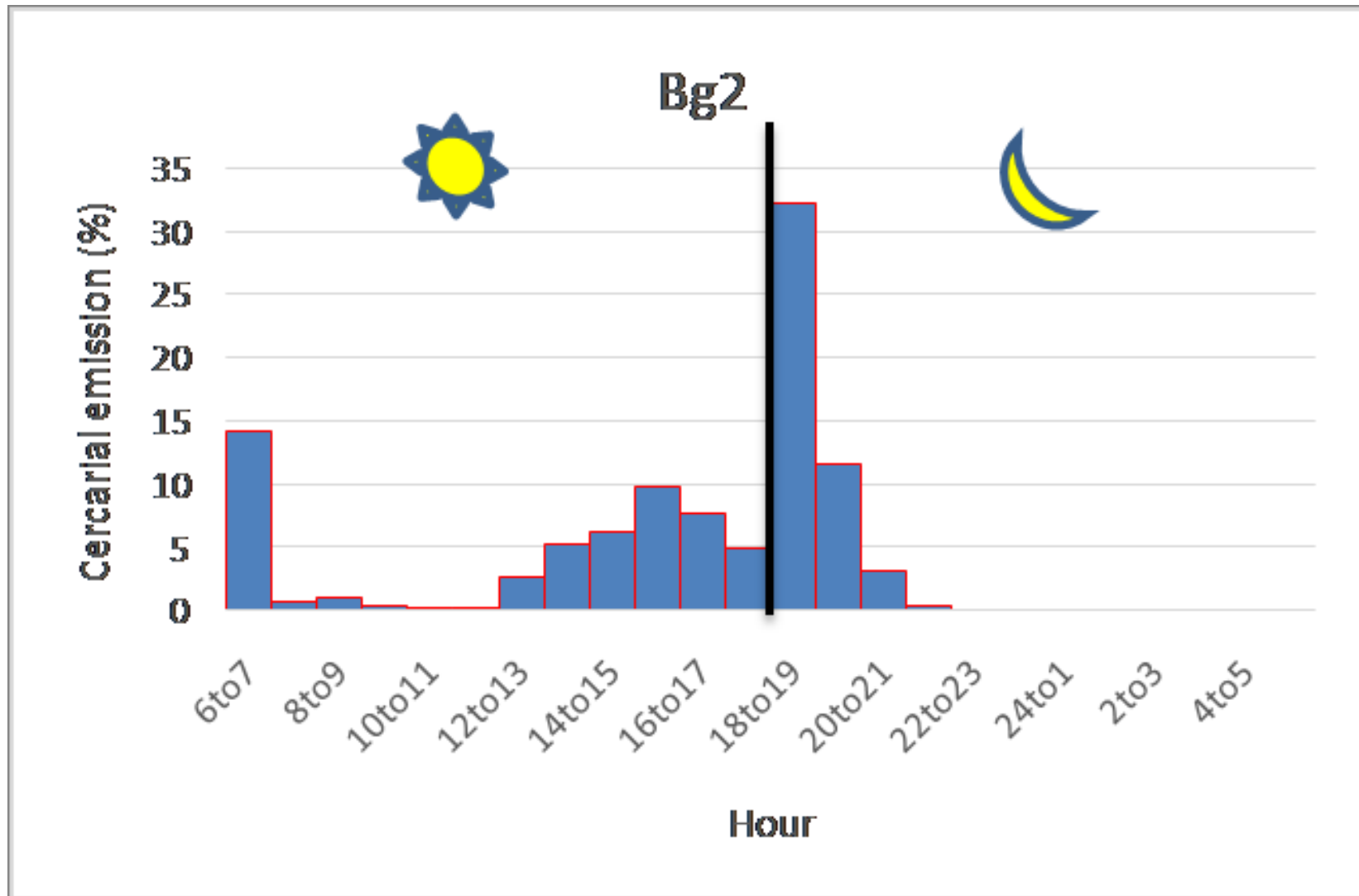
Experimental design:

- “ Snail exposure to **1 miracidium** from **1 egg** collected from the liver and duodenum of *Mastomys natalensis*.
- “ Study of the **hourly** cercarial emission pattern during over 24 hours during **5 consecutive days**.
- “ Molecular characterization of cercariae from infected snails.

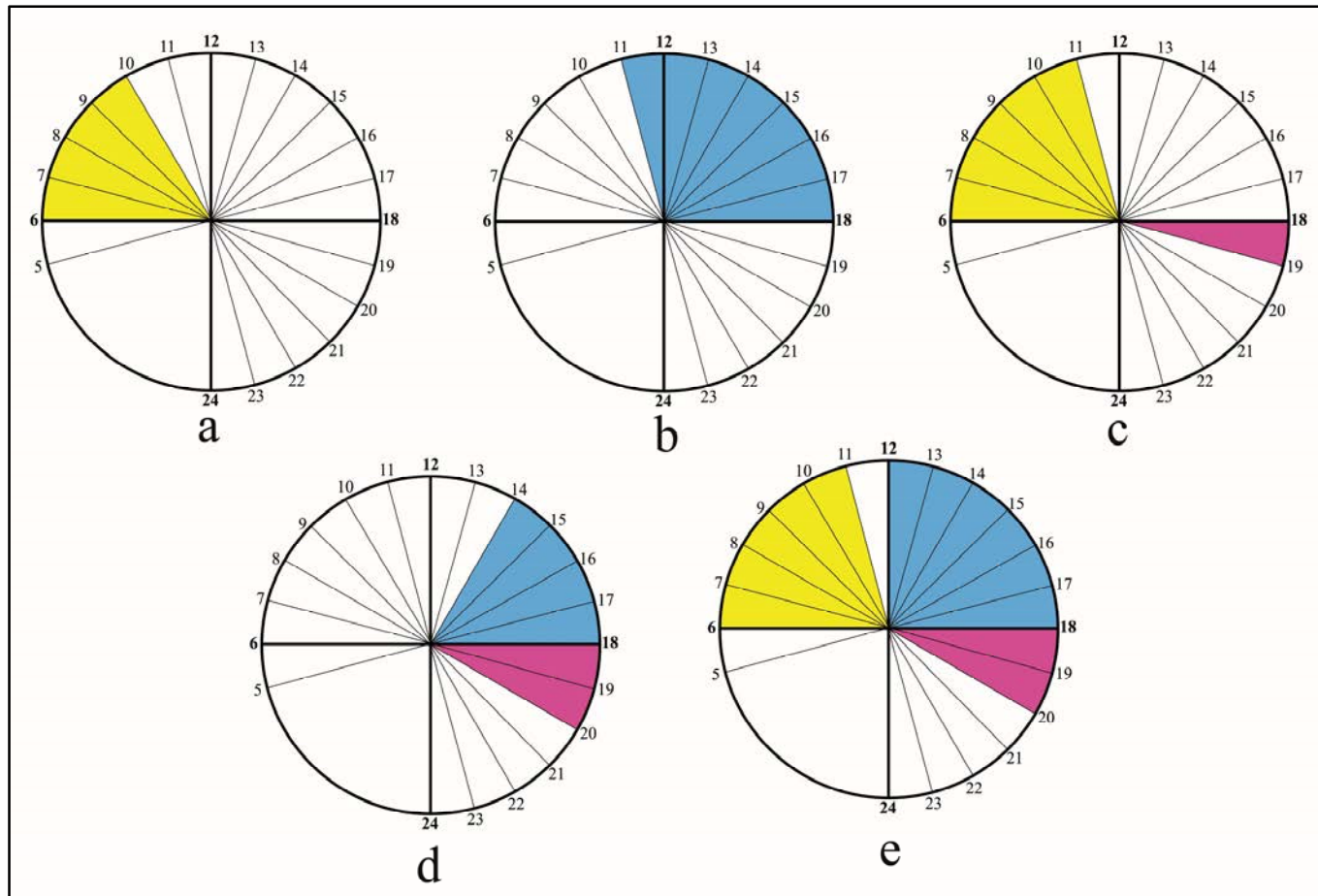




Late diurnal and nocturnal pattern observed in Cow schistosomes is present in *Mastomys natalensis* schistosomes.

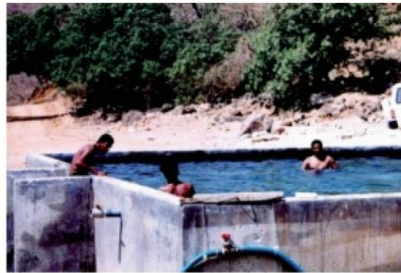
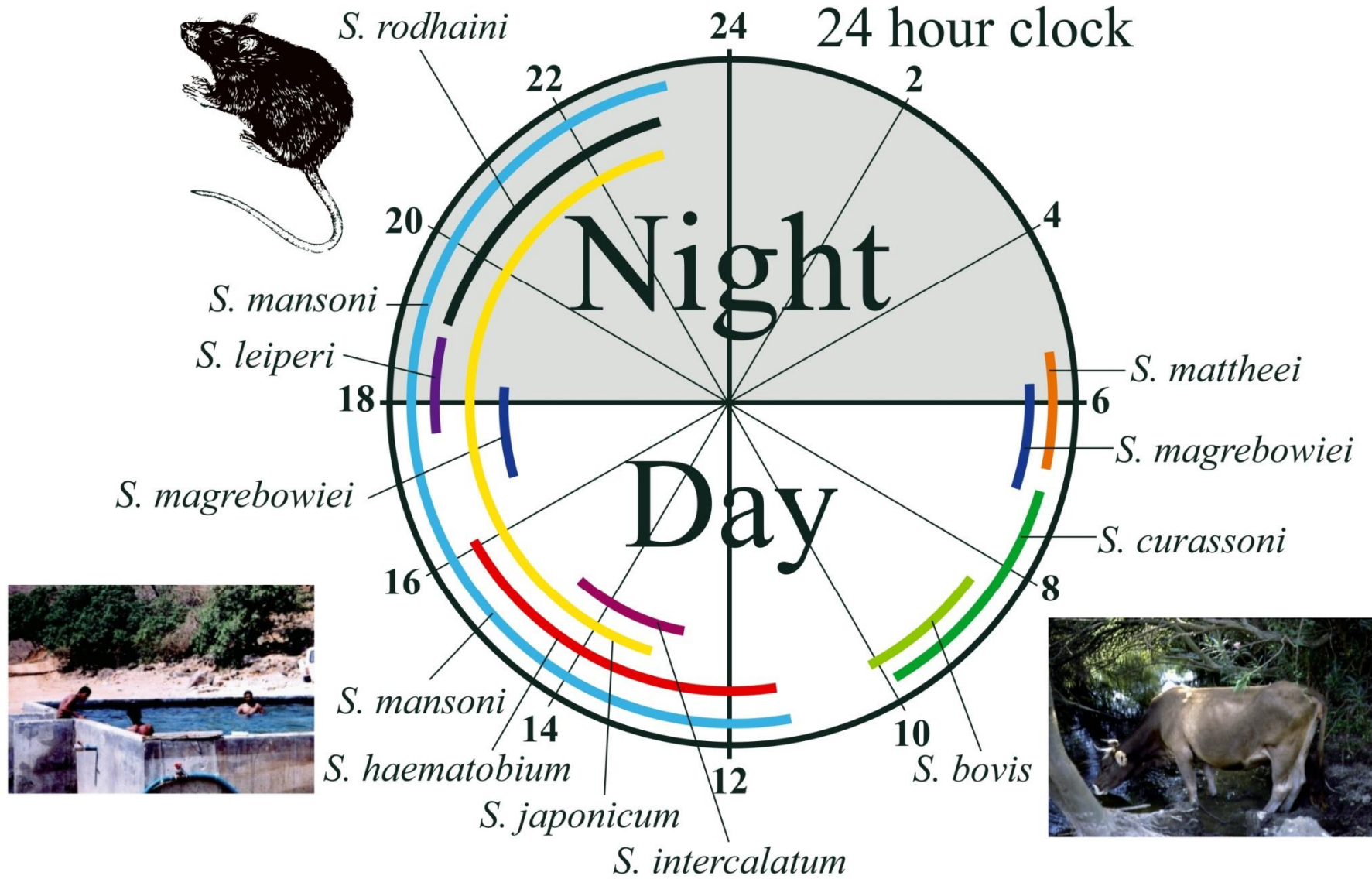


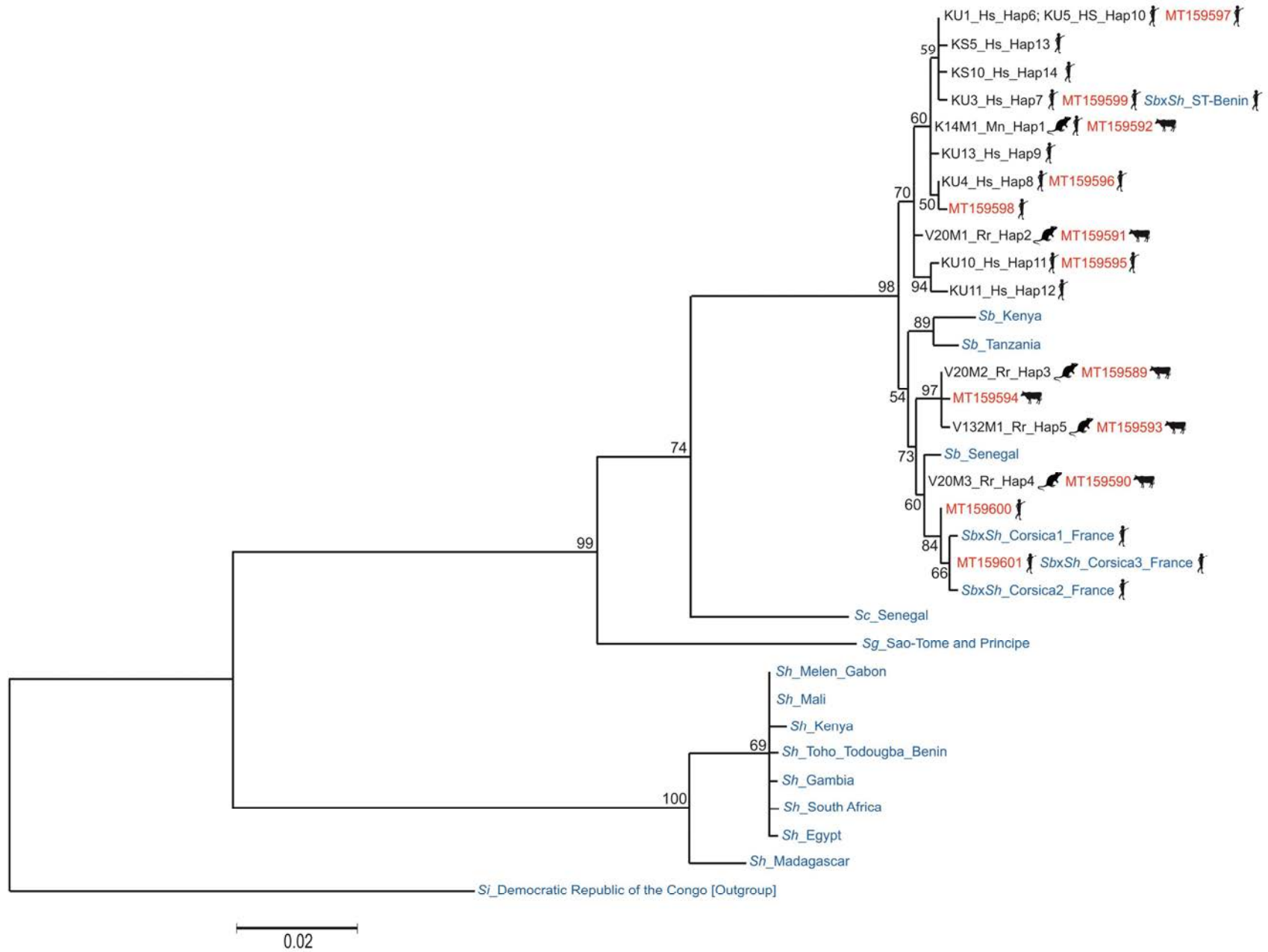
New cercarial emission pattern obtained in *Mastomys natalensis* schistosomes : **early, late and nocturnal pattern.**

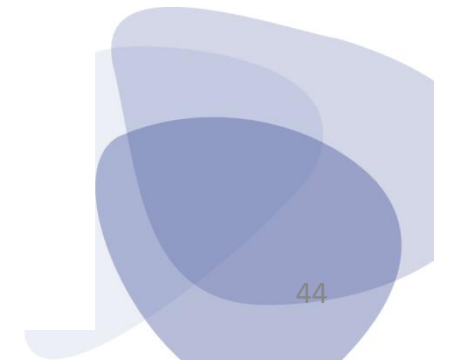
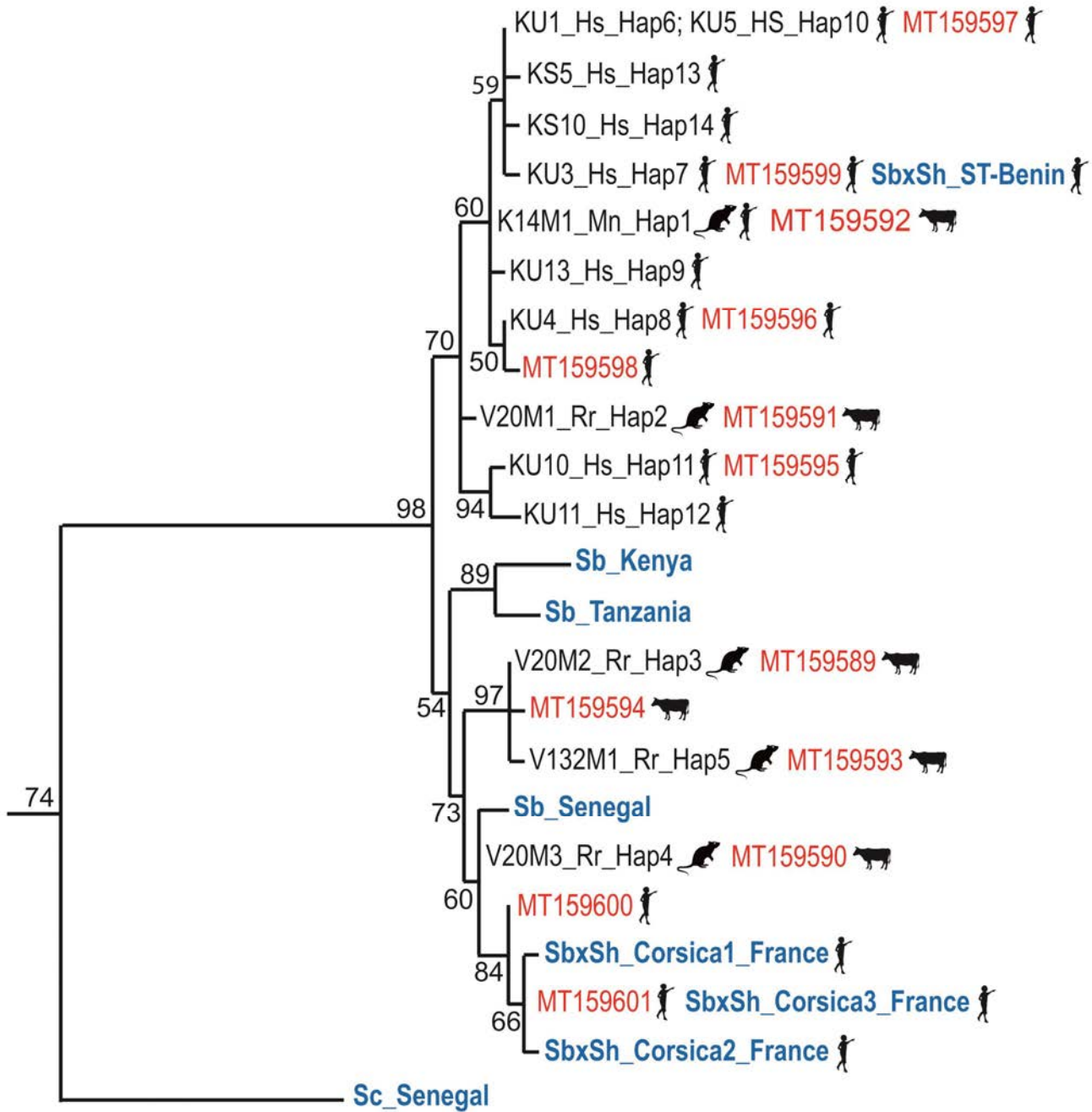


Five different patterns identified:

- **2** patterns with one peak each (in yellow typical pattern of *S. bovis* and in blue typical pattern of *S. haematobium*).
- **3** patterns that result from the combination with two or three peaks depending on the combination. In purple, nocturnal emission.







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Cotonou. Dr André Garcia

ANR CHRONOGET

Dispositif de Soutien aux
Collaborations avec l'Afrique
subsaharienne
AAP_2021 du CNRS

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Biologiques; Ecole d'Abomey-Calavi, Université d'Abomey-Calavi, Benin



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Merci

