Adaptive population differentiation in the blood fluke *Schistosoma*

Philip Louise M. Salvo

pmsalvo@up.edu.ph

Introduction

The differentiation of populations can be seen as incipient speciation events, giving us insight into the origin of species. Parasites are ideal models for evolutionary processes, especially selection, due to their interaction with the host and their modes of transmission having high potential for diversification and specialization (Huyse et al., 2005). Parasites of the trematode genus *Schistosoma* cause the Neglected Tropical Disease schistosomiasis. *Schistosoma* species require a mammalian/avian definitive host wherein the adults reside and sexually reproduce, and a snail intermediate host where larval stages asexually reproduce. In 2021, an estimated 251.4 million people worldwide were infected with schistosomiasis, inducing anemia, stunted growth, hypertension, impaired cognition, inflammation, and organ scarring (Colley et al., 2014; WHO, 2023). This paper is a compilation of research data on the emergence of distinct *Schistosoma* populations through the evolution of a unique adaptative phenotype, focusing on the selection of advantageous traits or genes on the three main species that infect humans: *Schistosoma japonicum, S. mansoni*, and *S. haematobium*.

1. Schistosoma japonicum

Schistosoma japonicum causes hepatic and intestinal schistosomiasis (Yang et al., 2023). There are 5 extant populations located in different regions: the Chinese lake/marshland region, the Chinese mountainous region, Taiwan, the Philippines, and the Indonesian island of Sulawesi (Luo et al., 2022). There was also a population from Japan that was declared extirpated in 1996 (Minai et al., 2003). The Chinese lake/marshland population was most likely the original population from which the other populations dispersed (Luo et al., 2022).

1.1 Adaptations by the Taiwan population

The Taiwan population was the first to diverge from the rest. It was estimated by Luo et al. (2022) that this population diverged ~45,000 years ago, coinciding with the divergence of the Taiwanese snail host *Oncomelania hupensis formosana* from the southeastern Chinese snail host *O. h. tangi*. The Taiwan schistosome is the only *S. japonicum* population that can infect the Taiwan snail. The divergence of the Taiwanese *S. japonicum* occurred prior to the arrival of humans in Taiwan. Consequently, the Taiwan lineage of *S. japonicum* is adapted to being zoophilic, and is unable to successfully develop in humans. It is also the only population that is hypothesized to have dispersed together with its snail intermediate host rather than being dispersed by humans during the spread of rice agriculture (Yin et al., 2015; Luo et al., 2022).

The Taiwan population's distinct intermediate and definitive host preferences are a likely cause for differential selection of genes involved in invasion, development, and colonization in the definitive host. Luo et al. (2022)'s cross-population analyses between the genomes of the Taiwan and the Chinese Lake populations revealed 499 selected coding genes in the Taiwan lineage. The study's within-population analysis for Taiwan detected 311 selected coding genes. Most of the selected genes were involved in DNA damage response, N-glycan processing, histone H2A acetylation, zinc ion transport, and embryonic development. This suggests that these gene categories play key roles in schistosome development, reproduction, or survival in different environments and hosts. A gene with a strong selection signal in Taiwan is the transcription repressor GATA Zinc Finger Domain

Containing 2A (*GATAD2A*), a gene required for stem cell pluripotency which promotes schistosome growth, survival, and reproductive organ development. The Taiwan *GATAD2A* is highly differentiated from the rest of *S. japonicum*, with four completely differentiated nucleotide sites. One fixed C>A Single Nucleotide Polymorphism (SNP) in the 5′ untranslated region possibly affects translation of the protein product. Another selected gene is Taiwan is the *CD63* gene, which was found to have a strong selection signal in within-population analyses. This gene is most highly expressed in the cercaria and encodes a tetraspanin CD63 receptor associated with development, maturation, and immune evasion in the definitive host. F_{ST} analysis identified *p40* as another selected gene in Taiwan. It encodes a major egg antigen that induces a strong Th1-polarized immune response in definitive hosts. A less antigenic variant would help in immune system evasion and reduce fibrosis in the host's liver (Luo et al., 2022).

In the Taiwan Z chromosome, the *DYS, LCAT,* and *Hmcn1* showed positive selection. *DYS* codes for dystrophin, a gene expressed in the muscles and nerves, and is associated with motor system homeostasis and regeneration. LCAT codes for lecithin-cholesterol acyltransferase, which converts cholesterol from the host bloodstream to the cholesteryl ester needed by the parasite for female reproductive organ development and egg embryonation. *Hmcn1* encodes HEMICENTIN 1, a gene associated with gonad development. It is highly expressed in sporocysts and adult males, suggesting function in both intermediate and definitive hosts (Zhou et al., 2024). These selected genes in the Taiwan population may manifest phenotypically as the Taiwan strain's weaker pathology with smaller hepatic granuloma, longer incubation period, longer time until host death, smaller eggs, different embryonated egg shape, variable number of testes, irregular testes arrangement, multiple cecal loops, smaller cercaria, smaller hepatic granuloma, and inability to successfully colonize humans (He et al., 1996; Fan, 2006; Luo et al., 2022). Overall, the Taiwan population seems to have traded resource exploitation and development rate for a weaker pathology, possibly adapting to a "slow and steady" strategy to prolong the life of the Taiwan fluke's animal definitive hosts.

1.2. Adaptation to the Chinese Mountain region

The two Chinese populations are geographically divided, with the Mountain population in the southwestern provinces of Sichuan and Yunnan, and the Lake/Marshland population in the eastern provinces. These two *S. japonicum* populations differ in their compatibility with the snail subspecies. The Chinese Mountain parasites can infect both the mountain and lowland variants of Chinese *O. hupensis* snail. The Chinese Lake parasites, however, can infect only the lowland snail variant but not the mountain variant (He et al., 1991). Given that the mountain snail subspecies is an outgroup of the lowland snails that diverged 2-6 million years ago, while the Mountain fluke diverged from the Lake population only ~5,400 years ago (Zhao et al., 2010; Luo et al., 2022), this suggests that instead of mountain co-evolution, the Mountain fluke only relatively recently adapted to also infect the mountain snail. Indeed, it is believed that the Mountain population descends from schistosomes carried by rice-planting humans expanding from the middle or lower reaches of the Yangtze River upstream to the Mountain region during the Neolithic agriculture era (Yin et al., 2015). Yin et al. (2016) found unique genetic markers in the Mountain population by comparing homozygous SNPs between pooled worms from 1 site from the Mountain and 3 sites from the Lake population. In the study, the Mountain population and a Lake site subpopulation from Guichi, Anhui formed phylogenetic sister branches, indicating that the ancestors of the Mountain population likely dispersed from Guichi or a nearby location. The Mountain lineage was found to have accumulated more mutations than its sister branch. There are 66 genes, involved mainly in nitrogen metabolism, with unique genetic changes in the Mountain site compared to the other sites. These genes may affect the efficiency of energy uptake and assimilation systems. Most of the enriched metabolic processes generate energy for the regulation of Rab/Ras GTPase activities, which are responsible for the regulation of signaling, especially in rapid response to environmental stimuli (Yin et al., 2016).

Selection signals from the Mountain fluke's adaptation to the Mountain snail host or environment were also investigated by Luo et al. (2022) using cross-population analysis between the Mountain and Lake populations. A total of 543 candidate genes were detected, mostly associated with neuronal stem cell population maintenance, protein-coupled receptor activity, and

metallopeptidase activity. The membrane-bound metallopeptidase Leishmanolysin (*Lmln*) gene showed significant positive selection in the Mountain population (Luo et al., 2022). The Lmln protein of schistosome sporocysts interferes with the immune cells of the snail intermediate host (Hambrook et al., 2018). Four non-synonymous mutations near the active pocket of Lmln are almost fixed in the mountain population but are rare in the lake/marshland (Luo et al., 2022). These mutations possibly make the Mountain lineage more adapted to evading *O. h. robertsoni* immune cells, which the Lake flukes likely cannot do.

In the Z chromosome, 266 candidate genes were identified, which were predominantly associated with protein serine/threonine kinase activity, metallo-endopeptidase activity, and dynein light chain binding (Zhou et al., 2024). The *Rab6* gene was noted as likely to be related to the unique compatibility of the Mountain parasite with the Mountain snail. This gene codes for a Ras-related protein that is an evolutionarily conserved small GTPase. *Rab6* is involved in regulation of retrograde transport in the Golgi body, cell mitosis, innate immunity, and epidermal integrity (Zhou et al., 2024). This gene's activity is likely one of the Ras/Rab GTPase activities that are being maintained by the metabolic processes found to be naturally selected by Yin et al. (2016). In *S. japonicum*, *Rab6* was more highly expressed in the miracidia. Its natural selection in the Mountain fluke might be adaptations for locating the Mountain snail host, stress responses to the Mountain climate, and protection against hyperparasites in the Mountain region that may infect the fluke. Another selected gene that may be involved in locating the preferred snail host is *VCP*, a gene that encodes Valosin-containing protein, which is an AAA+ ATPase. It also has function in germ cell development and immune evasion in the mammal host. It is highly expressed in the sporocyst stage, suggesting an important unknown function in the snail host. Another selected Z chromosome gene, *Nep4,* encodes the metallopeptidase Neprilysin4 (Zhou et al., 2024). Similar to the autosomal metallopeptidase *Lmln*, it protects the parasite from the snail immune system. In schistosomes, neprilysin, also called neural endopeptidase, is involved in the production of immunoactive peptides, which could inactivate the immunocytes from the snail host (Duvaux-Miret et al., 1992).

Positive selection in the Mountain region has also been found in the tegument-associated antigenic gene *SjT22.6*, detected from the high ratio of nonsynonymous/synonymous mutations. The positively selected allele, referred to as MHap is found only in the Mountain region, and is the most common haplotype (12 out of 14) in the mountainous Sichuan province. It is very differentiated from the other *SjT22.6* haplotypes, being 15 nucleotides separated from the closest other haplotype. As an antigenic gene, it is an immune response target, which has likely driven the rapid mutation of this haplotype (Li et al., 2017). Like the other selected genes in the Mountain region, the positive selection of this gene is likely caused by adaptation to the Mountain snail or environment.

1.3. Adaptations within the Chinese Lake/Marshland population

Within the *S. japonicum* Lake/Marshland population, there is a subpopulation in the Anhui province with distinct adaptive characteristics. The Anhui province is in eastern China, far away from the mountainous region of the southwest, but there is a hilly region in the southern part of the province where a subpopulation has been found with a late afternoon/evening (5:00-7:00 PM) cercaria shedding time (Lu et al., 2009; Su et al., 2013). This is in contrast with the Anhui marshland subpopulation, which like the rest of the Lake population, has a morning (7:00-11:00 AM) cercaria shedding time ((Lu et al., 2009; Su et al., 2013). It has been suggested that these shedding patterns are adaptations to increase chances of infection of their respective definitive hosts, as diurnal bovines are the main reservoir in the Anhui marshland, while nocturnal rodents are the reservoir in the Anhui hilly area (Lu et al., 2009; Lu et al., 2010). Nocturnal shedding times have also been observed in the Philippine population and the now extirpated Japanese population of *S. japonicum* (Kawashima et al., 1985). However, it is not known whether these are schistosome adaptations or a consequence of having *O. h. quadrasi* and *O. h. nosophora*, respectively as intermediate hosts. The Anhui subpopulations, however, have the same snail host *O. h. hupensis.* Additionally, the Anhui hill fluke has a night/late afternoon shedding time in both marshland and hill snails, suggesting that cercarial emergence time depends on the schistosome genetics, not the snail (Su et al., 2013).

Sun et al. (2023) sequenced the whole genomes of 4 Anhui hill flukes and 2 Anhui marshland flukes to search for selection signals. There was high

number of nonsynonymous SNPs. The marshland flukes had 276 selected genes, but none were significantly enriched. The hill flukes only had 78 selected genes, but 12 were significantly enriched. The paper concluded that the marshland subpopulation had more selected genes because they are subjected to higher priority control interventions due to bovines and humans being key hosts. None were enriched, however, because the control efforts are relatively recent. The hill subpopulation had fewer selection pressures because the rodents are not able to be targeted by control efforts due to logistic difficulties. However, they had many enriched genes because they have been adapting to a rodent host for a long time. The 12 enriched genes are mainly related to transcription and intercellular signal transduction such as zinc finger protein and members of the Mothers Against Decapentaplegic (SMAD) protein family. It has been implied that expression of SMAD is involved in the parasite's exploitation of the mammalian host's own key signaling pathways and growth factors as development signals for the schistosome. Other enriched genes are the epigenetic modifier histone-lysine N-methyltransferase and a transcription factor partner of Notch receptor. Notch signaling in sensory structures may play a role in the hill fluke's nocturnal cercarial emergence (Sun et al., 2023).

Within the Anhui marshland subpopulation, Wang et al. (2006) found host-dependent genetic differentiation between flukes from water buffalo, cattle and humans, and those from goats, pigs, dogs and cats. However, there is not enough evidence to conclude whether this is due to sympatric strain divergence or differential transmission between host species.

1.4. Possible adaptations in the Philippines

Though evidence is insufficient, a possible selection signal has been identified in the Philippine province of Leyte. In Moendeg et al.'s 2017 study of the differentiation of *S. japonicum* between Philippine provinces, they noted a possible adaptative differentiation in the Leyte subpopulation. In their study, allelic richness is usually positively associated with human prevalence. An exception is the *S. japonicum* from Leyte province, which had the lowest allelic richness despite having a relatively high human infection prevalence. The authors suggested that this could be due to the prolonged mass administrations of the antihelminthic drug Praziquantel (PZQ) in the province

(Moendeg et al., 2017). Leyte is one of the oldest endemic foci in the Philippines, as well as the site of the first clinical trial for PZQ in the late 1970s and the first province-wide PZQ chemotherapy program in 1982 (Inobaya et al., 2015; Leonardo et al., 2016). Though low allelic richness by itself may just be a consequence of a bottleneck caused by elimination efforts, the relatively high human prevalence possibly suggests a fitness advantage in the Leyte subpopulation, which may be caused by PZQ resistance. Low allelic diversity has previously been associated with drug resistance in schistosomes and other parasites (Coeli et al., 2013; Vianney et al., 2022; Early et al., 2025).

It speculated by authors that the Philippine strain of *S. japonicum* is more adapted to dogs and rats than the Chinese marshland strain (Carabin et al., 2015; Rudge et al., 2008). In Samar, *S. japonicum* from humans shared many genotypes with conspecifics from dogs, suggesting high dog-human transmission (Rudge et al., 2008), while in the Chinese marshland, *S. japonicum* genotypes from humans clearly clustered only with those from water buffalos and cattle and away from dogs (Wang et al., 2006). Additionally, a late afternoon/evening cercarial emergence has been found in Leyte *S. japonicum* (Kawashima et al., 1985), which is similar to the emergence time of the Anhui hilly subpopulation with dog and rodent reservoir hosts (Lu, Rudge, et al., 2010; Su et al., 2013). Nevertheless, water buffalos still remain significant reservoirs due to their large volume of fecal output and high infection prevalence (Jiz et al., 2021).

1.5. Diversification in antigenic genes

Parasite antigenic genes diversify as a response to selection pressures imposed by the host immune system (Weedall & Conway, 2010). Tegument-associated surface proteins such as tetraspanins are especially prone. The tetraspanins CD63, Sm29-like, SjTSP2, SjTSP8, and SjTSP25 were found to have high interprovince variation in China, especially in their surface-exposed extracellular loop regions (Luo et al., 2022; Parsons et al., 2023; Young et al., 2015). Parsons et al. (2025) investigated the diversity among 3 *S. japonicum* antigen coding genes: tetraspanin 23 (TSP23), venom allergen-like protein 7 (VAL-7), and tegument allergen-like protein 1 (TAL-1). Despite extensive haplotype sharing, the study found host-specific antigen variations between *S. japonicum* from humans, dogs, cats, and pigs, suggesting adaptation to certain host species (Parsons et al., 2025).

2. *Schistosoma mansoni*

This species is endemic in 54 countries across Africa, the Middle East, South America and the Caribbean (Chitsulo et al., 2000). It causes intestinal and hepatic schistosomiasis (Elbaz & Esmat, 2013).

2.1. Adaptation to human hosts

It is hypothesized that the split between the sibing species *S. mansoni* and *S. rodhani* occurred as *S. mansoni* adapted to infecting humans during the rise of fishing communities ~126,500 years ago in East Africa. Comparison of nonsynonymous mutations between the whole genomes of the sibling species revealed 767 positively selected coding sequences in *S. mansoni*, with enrichment of genes for extracellular matrix structure proteins expressed during the schistosomula stage. A cercarial elastase and a cercarial allergenic protein were also significantly positively selected. All of these point to positive selection in genes involved in human infection or immune evasion. Additionally, 273 genes were under balancing selection, with enrichment of genes associated with cell junction proteins, cell adhesion molecules, and G-protein modulators. Other genes under balancing selection include immunoglobulin I-set domains, four dynein domains, and hsp40 (Crellen et al., 2016a). Balancing selection in parasites usually occur in antigenic genes to avoid recognition by the host immune system.

2.2. Dispersal to West Africa and the Americas

S. mansoni was dispersed from East Africa to West Africa 6-7 thousand years ago, and then to the Americas through the Trans-Atlantic Slave trade (Crellen et al., 2016a). The West African population also gave rise to the Egyptian and Middle-eastern populations (Webster et al., 2013). It is expected that these migrations would cause selection on the dispersed populations as they adapted to the new environments and new *Biomphalaria* snail host populations. Adaptations by the dispersed populations may include their shorter prepatent period and higher virulence compared to the East African population (Panitz, 1966). Platt II et al. (2022)'s study examined selection in these genomes and found 4 uniquely selected genomic regions in Brazil, encompassing 80 candidate genes. Among them, 9 had very strong selection signals. Several of these genes are associated with housekeeping functions, including transcription and protein degradation. These selected

genes may have been involved in adaptation to the new environment or the new intermediate snail host species in Brazil. This adaptation is apparent, as African *S. mansoni* strains cannot infect the Brazilian *Biomphalaria* snail hosts (Panitz, 1966). There were 3 selected genomic regions each from Niger and Senegal with one shared region, making a total of 5 regions from West Africa. The selected regions had 112 and 157 genes, respectively. The role of the uniquely selected genes in West Africa are currently unclear, though they may be adaptations to the environment or local snail hosts. There were no selected genomic regions found from Tanzania in East Africa.

2.3. Cercarial emergence of S. mansoni

Cercarial emergence in *S. mansoni* is usually at mid-day, likely to exploit the times at which humans bathe or drink water. The mid-day emergence has been found in Puerto Rico, South Africa, Sudan, and Saudi Arabia (Pitchford et al., 1969; Tameim et al., 1985; Zahed, 1992). However, various interactions with hosts have resulted in the evolution of a diversity of emergence rhythms. In Oman, *S. mansoni* collected from humans have a diurnal cercarial emergence (~11:00 AM), while those from rats have a nocturnal emergence (~8:00 PM) (Mouahid et al., 2012). In Benin, there is a primary peak at noon, and secondary peaks at dawn and dusk when the fishermen would be in contact with the water (Ibikounle et al., 2012). In the Guadeloupe archipelago in the Caribbean, there are three emergence rhythms for *S. mansoni* attributed to alternative alleles at a single locus. The early peak (11:00 AM) is most common at urbanized foci where humans are the definitive host. The late peak (4:00 PM) is common at the forest, hosted by the crepuscular rats (Théron, 1984). An intermediate peak (1:00 PM) representing the heterozygote was most common in the mangrove swamp where both humans and rats are hosts (Théron & Combes, 1988). The Brazilian fluke strain also had an afternoon peak (2:00-5:00 PM), but was controlled by a different locus from the Guadeloupe strain. Knowledge of *S. mansoni* cercarial emergence time has been used to guide schistosomiasis control efforts. A significant reduction in schistosomiasis prevalence was observed in canal cleaners for the Gezira irrigation project in Sudan after their working hours were changed so that they left the water before 10:00 AM, avoiding the peak emergence time at noon (Tameim et al., 1985).

An *S. mansoni* strain from Belo Horizonte, Southeast Brazil shed 8 times more cercaria and was more fatal to the snail than a strain from East Brazil (Le Clec¼h et al., 2019). It was concluded that these are "boom and bust" strategy adaptations by the Southeast Brazil strain to a high transmission site with intense intraspecific competition between *S. mansoni* individuals and interspecific competition between *S. mansoni* and other *Biomphalaria*-infecting trematodes in Belo Horizonte such as *Cyclocoelum mutabile*, echinostomatids, strigeids, derogenids, and clinostomatids (Assis & Pinto, 2024; De Souza et al., 1998). Nematodes may have also contributed to competition in *Biomphalaria glabrata* (Mougeot et al., 1977), however, no data was found on the nematodes infecting snails in Belo Horizonte. In contrast to the Southeast Brazil strain, the East Brazil strain is in a low transmission site with less competition and thus adapted the "slow and steady" strategy (Le Clec¼h et al., 2019). Linkage disequilibrium analysis by Le Clec'h et al. (2020) identified 3 major and 2 minor quantitative trait loci (QTLs) for the cercarial production trait. The major QTL on chr. 1 includes peptidylprolyl isomerase (PPIase) which regulates intracellular signaling, transcription, inflammation, immunomodulation, and apoptosis. On the other major QTL on chr. 3, there is G-protein coupled receptor kinase 2, involved in cell migration. On another major QTL on chr. 5, there is the Calmodulin IQ domain protein, a calcium sensor that can stimulate changes in the actin cytoskeleton. In the two minor QTLs, the top candidate genes encode a nucleoporin and a Ly-1 antibody reactive (LYAR) gene encoding a cell growth regulating nucleolar protein. Potential mechanisms for the QTLs were discussed by the authors. The nucleoporin, G-protein, and LYAR genes are likely involved in the clonal reproduction of the sporocysts. The Calmodulin and LYAR genes are likely candidates for control of the differentiation of sporocyst cells into cercaria. PPIase and G-protein are speculated to be involved in immunomodulation of snail immune cells (Le Clec'h et al., 2021a).

2.4. Compatibility of S. mansoni *with snail hosts*

Similar to *S. japonicum*, populations of *S. mansoni* are known to adapt to infect certain snail populations, such that only the Egyptian *S. mansoni* strain can infect Egyptian *Biomphalaria boissyi* snails, and American *S. mansoni* strains are much more successful than African strains in infecting American snails (Panitz, 1966). Ibikounlé et al. (2012) showed that *S. mansoni* has a shorter

prepatent period and is less likely to kill its intermediate host when the snail is from a sympatric population. In the Americas, *S. mansoni* strains are more compatible (higher infectivity rate) to the sympatric population of *Biomphalaria glabrata* than to an allopatric population (Galinier et al., 2017). This specific compatibility polymorphism has been attributed to the subset of *S. mansoni* Polymorphic Mucins (SmPoMucs) that the fluke strain expresses, as well as its expression levels. Perrin et al. (2013) discovered that the expressed subset of SmPoMucs isn't dependent on genotype, but on the DNA methylation status of the promoter regions. The study also discovered ancestral SmPoMucs gene duplications from the common ancestor of the American strains, increasing diversity of these mucins compared to the African strains. Infection of Brazilian *S. mansoni* strains on sympatric and allopatric (Guadeloupean) *B. glabrata* by Roquis et al. (2016) revealed five different histone methylation states, with the majority of epimutations occurring on transcribed regions in cercaria. Spontaneous epimutations regardless of host occurred at 64 schistosome genomic sites, with 45% of the epimutations being mitotically inherited. This has significant implications on schistosome adaptation. The differentiation of adaptive phenotypes may not only be due to selection, but also epigenetic modifications that can be acquired, reversed, or maintained through generations.

2.5. Drug resistance in S. mansoni

In the 90s, resistance to PZQ was found in *S. mansoni* isolates from villagers in Egypt, where the drug has been used aggressively for more than 10 years (Magdi et al., 1999). Reduced susceptibility to PZQ has also been found in Senegal and Kenya (Fallon et al., 1995; Melman et al., 2009). In Uganda, a study by Crellen et al. (2016b) discovered that *S. mansoni* from the Mayuge district in the northern shore of Lake Victoria, which had undergone 5-9 rounds of mass drug administration (MDA), were more resistant to PZQ than schistosomes from the Tororo district, which only had one round of MDA. Berger et al. (2021) found 25 regions with extreme integrated haplotype scores in the Mayuge district encompassing 183 genes. A locus in chromosome 2 with high selection signals and reduced nucleotide diversity contained 4 sodium/potassium/calcium exchanger proteins. Missense, nonsense or splicing variants were found in 3 of the 4 genes. These channels could potentially counter the TRPM-mediated Ca^{2+} influx induced by PZQ. The

$TRPM_{PZQ}$ channel is the target of the drug praziquantel, but no selection signals were found in the gene for the $TRPM_{PZQ}$ channel itself. In chromosome 4, a selected region encompassing 11 genes included a gene predicted to encode an ATP-binding cassette (ABC) transporter *smdr1,* which could be a means of PZQ efflux (Berger et al., 2021).

In an island group at the north of Lake Victoria, Vianney et al. (2022) found that a region in chromosome 5 was highly differentiated between villages with standard and intensive MDA. This region had 25 protein coding genes including a gene associated with an ABC transporter and a gene with calcium-dependent functions. In cross-population selection analyses, the Z sex chromosome was enriched. Between pre- and post-treatment samples, the post-treatment had 107 selected genes, enriched with genes with roles in transferase activity, metabolic processes, and nucleotide salvage pathway. Between standard and intensive, the worms from villages with intensive MDA had 132 selected genes, enriched with toxin response genes (Vianney et al., 2022).

3. *Schistosoma haematobium*

Schistosoma haematobium is endemic in 53 countries across Africa and the Middle East (Chitsulo et al., 2000). This species causes urogenital schistosomiasis.

3.1. Local adaptation in Ghana

Local adaptation to the corresponding snail host has been found in this species in Ghana, where the *S. haematobium* "Ke strain" can infect *Bulinus truncatus rohlfsi*, but not *Bulinus globosus*, while the "Pokoasi strain" can infect the latter snail, but not the former (McCullough, 1959).

3.2. Cercarial emergence of S. haematobium

It is more light-sensitive than *S. mansoni*. In West Africa, there is a gradient of light sensitivity from the well-shaded forest in the south to the brightly-lit savannah in the north, manifesting as earlier to later (11:00 AM to 1:00 PM) cercarial emergence in the laboratory (N'Goran et al., 1997). The

midday emergence is an adaptation to humans bathing during the hottest part of the day (Mintsa-Nguéma et al., 2014). It has also been suggested that there is a "shadow response" in this species; sudden covering with shadows stimulate cercarial emergence as an adaptation to exploiting human activity making shadows in the water (N'Goran et al., 1997).

3.3. Adaptive introgression in S. haematobium

Genomic surveillance for selection in *S. haematobium* has mainly been focused on adaptive introgression. Adaptive introgression can occur after hybridization if certain introgressed genes increase in frequency via natural selection. The *S. haematobium* clade contains 8 other species, and their close phylogenetic relationship makes hybridization not uncommon (Landeryou et al., 2022).

Comparison of the genomes of *S. haematobium* populations from River Niger and the Zanzibar Archipelago by Platt II et al. (2019) revealed that 65% of the Nigerian population had mitochondria derived from *S. bovis*. All the Nigerian samples had 3-8% of their autosomal genome derived from *S. bovis* due to an introgression event ~240 years ago, while the Zanzibari samples had no *S. bovis* introgression. Cross-population analysis revealed 17 regions under directional selection between the two populations, with the strongest selection signals located in chromosome 5, overlapping with the greatest introgression frequency. Within this region is a leishmanolysin homolog called invadolysin, which is involved in mammalian host immune evasion (Platt II et al. 2019). Introgression in this locus was similarly found in Egypt, Mali, and Corsica (Rey et al., 2021a). It is hypothesized that the common ancestor of these populations of *S. haematobium* received *S. bovis* genes through hybridization, and selection favored the fixation of this gene in the population (Platt II et al., 2019). Hybridization/introgression between these two species has also been found in Cameroon, Senegal, Benin, Ivory Coast, Malawi, Gambia, Guinea-Bissau, Sudan, Ethiopia, Uganda, Kenya, and the French island of Corsica, though the introgression is always unidirectional from *S. bovis* to *S. haematobium* (Kincaid-Smith et al., 2021; Platt II et al., 2019; Rey et al., 2021a). The 2013 outbreak of urogenital schistosomiasis in Corsica has been discovered to be caused by a population with its mitochondrial genome and 23% of its nuclear genome derived from *S. bovis* and 77% of its nuclear

genome derived from *S. haematobium* (Kincaid-Smith et al., 2021). Hybrid vigor may have given this population the capacity for permanent endemicity in Corsica.

Deforestation in Cameroon has allowed the establishment of the snail *Bulinus trunculatus* and the subsequent hybridization of the invasive *S. haematobium* with the native *S. guineensis*, leading to outbreaks of urogenital schistosomiasis. Selection in the hybrids has been inferred by Landeryou et al. (2022) using outlier loci; those with over- or under-representation of alleles from one of the parental species. This has revealed 37 overrepresented loci from *S. guineensis* and 129 from *S. haematobium*, as well as 13 underrepresented loci from *S. guineensis* and 49 from *S. haematobium*. The largest proportion of the outlier loci encoded for proteins that localize in the cell membrane. There was excess introgression of tegument and antigenic genes such as Tegument-Allergen-Like (TAL) and Venom Allergen-Like (VAL proteins) from *S. haematobium,* suggesting that these genes improve the hybrids' infective success through evasion or combat of the host immune system. There was a consistent presence among the outlier loci of genes associated with Ca^{2+}-gated channels or voltage-gated Ca^{2+} channels, which could possibly suggest adaptation to praziquantel (Landeryou et al., 2022). Praziquantel resistance has never been officially observed in either wild or laboratory populations of *S. haematobium* (Summers et al., 2022).

4. Laboratory populations

The occurrence of selection on laboratory populations of parasites can give insights on natural selection that may be occurring on wild parasite populations.

4.1. Adaptation to laboratory hosts

Adaptation to a specific type of mammalian host has been observed in laboratory populations of schistosomes, which has implications on the effect of reservoir host identity on parasite infectivity in the wild. A Kenyan population of *S. mansoni*, which has humans as its natural definitive host, was isolated in 1968 and subsequently passaged through two mammalian hosts: baboons and mice (Fletcher et al., 1981). These hosts represent

nonhuman primates and rodents, the two significant types of reservoir host for *S. mansoni* (Catalano et al., 2018; Richards et al., 2019). It was later discovered that the baboon fluke population showed greater molecular polymorphism (6 polymorphic loci) than the mice fluke population (1 polymorphic locus) (Fletcher et al., 1981). LoVerde et al. (1985)'s experiments later confirmed that the decrease in allelic diversity was caused by host-induced selection, not genetic drift. Passaging the baboon fluke population through mice caused independent selection of the same alleles and allelic frequency trends as the population that had been passaged through mice for 12 years. The adaptation of a schistosome population to a mammalian host might also influences its infectivity to another mammalian host, as shown by the experiment by Taylor et al. (1977), wherein a laboratory population of the sheep-infecting fluke *Schistosoma mattheei* showed decreased infectivity to sheep after passaging through hamsters. These findings in the laboratory populations have implications on the zoonotic potential of field populations, as they suggest that the identity of reservoir hosts have consequences on the infectivity of the schistosomes to humans. Theoretically, maintenance in primate reservoir hosts maintains the allelic diversity, and therefore the zoonotic potential of the schistosomes, while maintenance in rodent reservoirs would lead to adaptation to rodents, decreased allelic diversity, and decreased zoonotic potential.

4.2. Experimentally-induced praziquantel resistance

Artificial selection for praziquantel resistance in the laboratory has been independently replicated multiple times using different parental populations of *S. mansoni*, producing PZQ-resistance lines in as little as 2 generations (Couto et al., 2011; Fallon & Doenhoff, 1994; Lotfy et al., 2015; Pinto-Almeida et al., 2015; Sanchez et al., 2019). Praziquantel resistance has also been induced in the laboratory for *S. japonicum* using a population from a marshland region in Hunan province (Liang et al., 2011). Selection via PZQ treatment of an *S. mansoni* laboratory population has revealed a genetic basis for resistance. The PZQ dose-response curves showed a 14-fold difference in PZQ response between the parental population and the PZQ-selected population. Marker-assisted selection of a SNP in $TRPM_{PZQ}$ associated with resistance further increased the difference in PZQ response to 377-fold (Le Clec'h et al., 2021b). Reanalysis of the genomes of the resistant population

using the improved *S. mansoni* v10 reference genome revealed that a single QTL in chromosome 3 is responsible for PZQ resistance (Chevalier et al., 2024). This single resistance QTL spans ~5.7 Mb containing 137 genes, including several candidate resistance genes such as three partial ABC transporters, a voltage-gated calcium channel subunit, the transcription factor *SOX13*, and the praziquantel target itself, $TRPM_{PZQ}$ (Le Clec'h et al., 2021b).

Future directions

Though it seems like the targeted surveillance of specific loci or genes seems to be the logical next step after identification of "selectable" trait loci in genomic studies, there is a dearth in research using these previously identified loci to detect selection on schistosomes. In my literature review, I found only Li et al. (2017), which found selection on an *S. japonicum* antigenic gene. There is a need for the development of primers for molecular markers in these selectable trait loci so that they may be used for surveillance of selection for drug resistance, host-switching, or other adaptive mechanisms in schistosome populations by researchers who cannot afford whole-genome sequencing. A possible source of polymorphic markers are introns, regions spliced out of mRNA, and are therefore mostly not subjected to positive or negative selective pressures on proteins. Introns are relatively freer to undergo neutral molecular evolution compared to translated regions (Loewenthal et al., 2022; Resch et al., 2007). Introns of taurocyamine kinase has been used in several other trematode species and may be used as a polymorphic marker in schistosomes (Saijuntha et al., 2018, 2020; Tantrawatpan et al., 2021, 2023). The practice of using introns as markers with the flanking exon regions as the primers, called EPIC (exon-primed intron-crossing), becomes increasingly easy as more reference genomes become available online (Blasco-Costa et al., 2016).

A better quality *S. mansoni* reference genome was recently assembled, wherein there was correction of certain sites in chromosome 3 that were previously incorrectly included in chromosome 2 (Chevalier et al., 2024). The 2 QTLs for PZQ resistance have now been discovered to be a single QTL located in chromosome 3. It is expected that rearrangements of the reference

genomes for *S. japonicum* and *S. haematobium* will follow to restore collinearity between homologous chromosomes.

Summary and Conclusion

Schistosoma populations can differentiate from each other in response to selective pressures from snail intermediate hosts, definitive hosts, and human control efforts. Dispersal of a population to a new location can be accompanied by adaptations to the new local hosts, which leaves signs of selection in their genomes. Signals of selection can be detected with genomic surveillance, including within-population and cross-population analyses. A common trend in population differentiation is the diversification of cercarial emergence times, which are usually adaptations to the time that the definitive host is in contact with water. Antigenic proteins are also commonly selected, particularly when the schistosome population is adapting to a different host. Host-induced epigenetic modifications possibly facilitate adaptation to hosts much faster than selection of genes. Selection has been observed in several genes related to praziquantel activity or efflux in villages subjected to several rounds of mass drug administration. Among closely related species, hybridization can increase fitness through selective introgression of specific advantageous genes which allows hybrids to exploit new conditions that the parental species could not. This phenomenon is common in *S. haematobium*. The reservoir host of a schistosome population matters not only in their proximity and transmission risk to humans, but also because specific host-induced adaptations can alter the population's zoonotic potential. As genomic studies discover loci that experience selection in response to pressures, we are given candidate loci for specific targeted surveillance in populations of interest. With the growing concerns about parasites' adaptations to the selective pressures exerted by human interventions (Moendeg et al., 2017; Nikolakis et al., 2022; Rey, et al., 2021b), particularly mass drug administrations, genomic surveillance of parasite genomes and targeted surveillance of identified loci to detect natural selection become increasingly important.

Philip Louise M. Salvo *is currently finishing his MS Biology degree at the Institute of Biology, College of Science, University of the Philippines Diliman with a scholarship from the Accelerated Science and Technology Human Resource and Development Program (ASTHRDP) by DOST. He had previously attained his BS Biology degree (Cellular and Organismal Biology) from the same university institution. His research consists of molecular detection, DNA barcoding, and molecular phylogeny of helminthic parasites.*

REFERENCES

Assis, J. C. A. de, & Pinto, H. A. (2024). The Natural Infection of Freshwater Snails with the Avian Air Sac Fluke, *Cyclocoelum mutabile* (Trematoda: Cyclocoelidae), in Brazil. *Diversity*, *16*(7), Article 7. https://doi.org/10.3390/d16070422

Berger, D. J., Crellen, T., Lamberton, P. H. L., Allan, F., Tracey, A., Noonan, J. D., Kabatereine, N. B., Tukahebwa, E. M., Adriko, M., Holroyd, N., Webster, J. P., Berriman, M., & Cotton, J. A. (2021). Whole-genome sequencing of *Schistosoma mansoni* reveals extensive diversity with limited selection despite mass drug administration. *Nature Communications*, *12*(1), Article 1. https://doi.org/10.1038/s41467-021-24958-0

Blasco-Costa, I., Cutmore, S. C., Miller, T. L., & Nolan, M. J. (2016). Molecular approaches to trematode systematics: 'Best practice' and implications for future study. *Systematic Parasitology*, *93*(3), 295–306. https://doi.org/10.1007/s11230-016-9631-2

Carabin, H., McGarvey, S. T., Sahlu, I., Tarafder, M. R., Joseph, L., De Andrade, B. B., Balolong, E., & Olveda, R. (2015). *Schistosoma japonicum* in Samar, the Philippines: Infection in dogs and rats as a possible risk factor for human infection. *Epidemiology and Infection*, *143*(8), 1767–1776. https://doi.org/10.1017/S0950268814002581

Catalano, S., Sène, M., Diouf, N. D., Fall, C. B., Borlase, A., Léger, E., Bâ, K., & Webster, J. P. (2018). Rodents as Natural Hosts of Zoonotic *Schistosoma* Species and Hybrids: An Epidemiological and Evolutionary Perspective From West Africa. *The Journal of Infectious Diseases*, *218*(3), 429–433. https://doi.org/10.1093/infdis/jiy029

Chevalier, F. D., Le Clec'h, W., Berriman, M., & Anderson, T. J. C. (2024). A single locus determines praziquantel response in *Schistosoma mansoni*. *Antimicrobial Agents and Chemotherapy*, *68*(3), e01432-23. https://doi.org/10.1128/aac.01432-23

Chitsulo, L., Engels, D., Montresor, A., & Savioli, L. (2000). The global status of schistosomiasis and its control. *Acta Tropica*, *77*(1), 41–51. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5633072/

Coeli, R., Baba, E. H., Araujo, N., Coelho, P. M. Z., & Oliveira, G. (2013). Praziquantel Treatment Decreases *Schistosoma mansoni* Genetic Diversity in Experimental Infections. *PLoS Neglected Tropical Diseases*, *7*(12), e2596. https://doi.org/10.1371/journal.pntd.0002596

Colley, D. G., Bustinduy, A. L., Secor, W. E., & King, C. H. (2014). Human schistosomiasis. *The Lancet*, *383*(9936), 2253–2264. https://doi.org/10.1016/S0140-6736(13)61949-2

Couto, F. F. B., Coelho, P. M. Z., Araújo, N., Kusel, J. R., Katz, N., Jannotti-Passos, L. K., & Mattos, A. C. A. (2011). *Schistosoma mansoni*: A method for inducing resistance to praziquantel using infected *Biomphalaria glabrata* snails. *Memorias Do Instituto Oswaldo Cruz*, *106*(2), 153–157. https://doi.org/10.1590/s0074-02762011000200006

Crellen, T., Allan, F., David, S., Durrant, C., Huckvale, T., Holroyd, N., Emery, A. M., Rollinson, D., Aanensen, D. M., Berriman, M., Webster, J. P., & Cotton, J. A. (2016). Whole genome resequencing of the human parasite *Schistosoma mansoni* reveals population history and effects of selection. *Scientific Reports*, *6*, 20954. https://doi.org/10.1038/srep20954

Crellen, T., Walker, M., Lamberton, P. H. L., Kabatereine, N. B., Tukahebwa, E. M., Cotton, J. A., & Webster, J. P. (2016). Reduced Efficacy of Praziquantel Against *Schistosoma mansoni* Is Associated With Multiple Rounds of Mass Drug Administration. *Clinical Infectious Diseases: An Official Publication of the Infectious Diseases Society of America*, *63*(9), 1151–1159. https://doi.org/10.1093/cid/ciw506

De Souza, C. P., Lima, L. C., Jannotti-Passos, L. K., Ferreira, S. S., Guimarães, C. T., Vieira, I. B., & Mariani Júnior, R. (1998). Lymnic snails from the microregion of Belo Horizonte, Minas Gerais, Brazil with an emphasis on parasite disease vectors. *Revista Da Sociedade Brasileira De Medicina Tropical*, *31*(5), 449–456.

Duvaux-Miret, O., Stefano, G. B., Smith, E. M., Dissous, C., & Capron, A. (1992). Immunosuppression in the definitive and intermediate hosts of the human parasite *Schistosoma mansoni* by release of immunoactive neuropeptides. *Proceedings of the National Academy of Sciences of the United States of America*, *89*(2), 778–781. https://doi.org/10.1073/pnas.89.2.778

Early, A. M., Pelleau, S., Musset, L., & Neafsey, D. E. (2025). Temporal Patterns of Haplotypic and Allelic Diversity Reflect the Changing Selection Landscape of the Malaria Parasite *Plasmodium falciparum*. *Molecular Biology and Evolution*, *42*(4), msaf075. https://doi.org/10.1093/molbev/msaf075

Elbaz, T., & Esmat, G. (2013). Hepatic and Intestinal Schistosomiasis: Review. *Journal of Advanced Research*, *4*(5), 445–452. https://doi.org/10.1016/j.jare.2012.12.001

Fallon, P. G., & Doenhoff, M. J. (1994). Drug-Resistant Schistosomiasis: Resistance to Praziquantel and Oxamniquine Induced in *Schistosoma Mansoni* in Mice is Drug Specific. *The American Journal of Tropical Medicine and Hygiene*, *51*(1), 83–88. https://doi.org/10.4269/ajtmh.1994.51.83

Fallon, P. G., Sturrock, R. F., Niang, A. C., & Doenhoff, M. J. (1995). Short report: Diminished susceptibility to praziquantel in a Senegal isolate of *Schistosoma mansoni*. *The American Journal of Tropical Medicine and Hygiene*, *53*(1), 61–62.

Fan, P.-C. (2006). The History of Schistosomiasis Japonica in Taiwan. *The Kaohsiung Journal of Medical Sciences*, *22*(7), 309–320. https://doi.org/10.1016/S1607-551X(09)70317-1

Fletcher, M., LoVerde, P. T., & Woodruff, D. S. (1981). Genetic Variation in *Schistosoma mansoni*: Enzyme Polymorphisms in Populations from Africa, Southwest Asia, South America, and the West Indies. *The American Journal of Tropical Medicine and Hygiene*, *30*(2), 406–421. https://doi.org/10.4269/ajtmh.1981.30.406

Galinier, R., Roger, E., Moné, Y., Duval, D., Portet, A., Pinaud, S., Chaparro, C., Grunau, C., Genthon, C., Dubois, E., Rognon, A., Arancibia, N., Dejean, B., Théron, A., Gourbal, B., & Mitta, G. (2017). A multistrain approach to studying the mechanisms underlying compatibility in the interaction between *Biomphalaria glabrata* and *Schistosoma mansoni*. *PLOS Neglected Tropical Diseases*, *11*(3), e0005398. https://doi.org/10.1371/journal.pntd.0005398

Hambrook, J. R., Kaboré, A. L., Pila, E. A., & Hanington, P. C. (2018). A metalloprotease produced by larval *Schistosoma mansoni* facilitates infection establishment and maintenance in the snail host by interfering with immune cell function. *PLOS Pathogens*, *14*(10), e1007393. https://doi.org/10.1371/journal.ppat.1007393

He, Y., Hu, Y., Yu, Q., Xie, M., & Qiu, L. (1996). Comparison of some biological aspects among seven geographic strains of *Schistosoma japonicum* in Asia. *Chinese Journal of Parasitology and Parasitic Diseases*, *14*(3), 212–218. https://www.jsczz.cn/EN/

He, Y. X., Guo, Y. H., Ni, C. H., Xia, F., Liu, H. X., Yu, Q. F., & Hu, Y. Q. (1991). Compatibility between *Oncomelania hupensis* and different isolates of *Schistosoma japonicum* in China. *The Southeast Asian Journal of Tropical Medicine and Public Health*, *22*(2), 245–248.

Huyse, T., Poulin, R., & Théron, A. (2005). Speciation in parasites: A population genetics approach. *Trends in Parasitology*, *21*(10), 469–475. https://doi.org/10.1016/j.pt.2005.08.009

Ibikounle, M., Mone, H., Abou, Y., Kinde-Gazard, D., Sakiti, N. G., Mouahid, G., & Massougbodji, A. (2012). Premier cas de chronobiologie des émissions cercariennes de type infradien chez *Schistosoma mansoni* dans deux foyers du sud-Bénin. *International Journal of Biological and Chemical Sciences*, *6*(3), Article 3. https://doi.org/10.4314/ijbcs.v6i3.14

Inobaya, M. T., Olveda, R. M., Tallo, V., McManus, D. P., Williams, G. M., Harn, D. A., Li, Y., Chau, T. N. P., Olveda, D. U., & Ross, A. G. (2015). Schistosomiasis mass drug administration in the Philippines: Lessons learnt and the global implications. *Microbes and Infection*, *17*(1), 6–15. https://doi.org/10.1016/j.micinf.2014.10.006

Jiz, M., Mingala, C., Fu, Z.-Q., Adriatico, M., Lu, K., Jarilla, B., Sagliba, M., Moreno, A., Park, S., Lin, J.-J., Olveda, R., Kurtis, J. D., & Wu, H. W. (2021). High prevalence of *Schistosoma japonicum* by perfusion in naturally exposed water buffalo in a region of the Philippines endemic for human schistosomiasis. *PLoS Neglected Tropical Diseases*, *15*(9), e0009796. https://doi.org/10.1371/journal.pntd.0009796

Kawashima, K., Blas, B. L., & Santos, A. T. (1985). The cercarial emergence of *Schistosoma japonicum* from *Oncomelania quadrasi* under outdoor conditions in the Philippines. *Journal of Helminthology*, *59*(3), 225–231. https://doi.org/10.1017/s0022149x00007987

Kincaid-Smith, J., Tracey, A., de Carvalho Augusto, R., Bulla, I., Holroyd, N., Rognon, A., Rey, O., Chaparro, C., Oleaga, A., Mas-Coma, S., Allienne, J.-F., Grunau, C., Berriman, M., Boissier, J., & Toulza, E. (2021). Morphological and genomic characterisation of the *Schistosoma* hybrid infecting humans in Europe reveals admixture between *Schistosoma haematobium* and *Schistosoma bovis*. *PLoS Neglected Tropical Diseases*, *15*(12), e0010062. https://doi.org/10.1371/journal.pntd.0010062

Landeryou, T., Rabone, M., Allan, F., Maddren, R., Rollinson, D., Webster, B. L., Tchuem-Tchuenté, L.-A., Anderson, R. M., & Emery, A. M. (2022). Genome-wide insights into adaptive hybridisation across the *Schistosoma haematobium* group in West and Central Africa. *PLOS Neglected Tropical Diseases*, *16*(1), e0010088. https://doi.org/10.1371/journal.pntd.0010088

Le Clec'h, W., Chevalier, F. D., McDew-White, M., Menon, V., Arya, G.-A., & Anderson, T. J. C. (2021). Genetic architecture of transmission stage production and virulence in schistosome parasites. *Virulence*, *12*(1), 1508–1526. https://doi.org/10.1080/21505594.2021.1932183

Le Clec'h, W., Chevalier, F., Mattos, A., Strickland, A., Diaz, R., McDew-White, M., Rohr, C., Kinung'hi, S., Allan, F., Webster, B., Webster, J., Emery, A., Rollinson, D., Garba, A., Mashikhi, K., Yafae, S., Idris, M., Moné, H., Mouahid,

G., & Anderson, T. (2021). Genetic analysis of praziquantel response in schistosome parasites implicates a transient receptor potential channel. *Science Translational Medicine*, *13*. https://doi.org/10.1126/scitranslmed.abj9114

Le Clec'h, W. L., Chevalier, F. D., McDew-White, M., Menon, V., Arya, G.-A., & Anderson, T. J. C. (2020). *Genetic architecture of transmission stage production and virulence in schistosome parasites* (p. 2020.12.23.424224). bioRxiv. https://doi.org/10.1101/2020.12.23.424224

Le Clec¼h, W., Diaz, R., Chevalier, F. D., McDew-White, M., & Anderson, T. J. C. (2019). Striking differences in virulence, transmission and sporocyst growth dynamics between two schistosome populations. *Parasites & Vectors*, *12*(1), 485. https://doi.org/10.1186/s13071-019-3741-z

Leonardo, L., Chigusa, Y., Kikuchi, M., Kato-Hayashi, N., Kawazu, S., Ma Angeles, J., Fontanilla, I. K., Tabios, I. K., Moendeg, K., Goto, Y., Fornillos, R. J., Tamayo, P. G., & Chua, J. C. (2016). Schistosomiasis in the Philippines: Challenges and some successes in control. *Southeast Asian Journal of Tropical Medicine and Public Health*, *47*(4). https://registry.healthresearch.ph/index.php/herdin-home?view=research&cid=71533

Li, Y., Yin, M., Wu, Q., McManus, D. P., Blair, D., Li, H., Xu, B., Mo, X., Feng, Z., & Hu, W. (2017). Genetic diversity and selection of three nuclear genes in *Schistosoma japonicum* populations. *Parasites & Vectors*, *10*, 87. https://doi.org/10.1186/s13071-017-2033-8

Liang, Y.-S., Li, H.-J., Dai, J.-R., Wang, W., Qu, G.-L., Tao, Y.-H., Xing, Y.-T., Li, Y.-Z., Qian, K., & Wei, J.-Y. (2011). Studies on resistance of *Schistosoma* to praziquantel XIII: Resistance of *Schistosoma japonicum* to praziquantel is experimentally induced in laboratory. *Chinese Journal of Schistosomiasis Control*, *23*(6), 605–610.

Loewenthal, G., Wygoda, E., Nagar, N., Glick, L., Mayrose, I., & Pupko, T. (2022). The evolutionary dynamics that retain long neutral genomic sequences in face of indel deletion bias: A model and its application to human introns. *Open Biology*, *12*(12), 220223. https://doi.org/10.1098/rsob.220223

Lotfy, W. M., Hishmat, M. G., El Nashar, A. S., & Abu El Einin, H. M. (2015). Evaluation of a method for induction of praziquantel resistance in *Schistosoma mansoni*. *Pharmaceutical Biology*, *53*(8), 1214–1219. https://doi.org/10.3109/13880209.2014.970289

Lu, D.-B., Rudge, J. W., Wang, T.-P., Donnelly, C. A., Fang, G.-R., & Webster, J. P. (2010). Transmission of *Schistosoma japonicum* in Marshland and Hilly Regions of China: Parasite Population Genetic and Sibship Structure. *PLOS Neglected Tropical Diseases*, *4*(8), e781. https://doi.org/10.1371/journal.pntd.0000781

Lu, D.-B., Wang, T.-P., Rudge, J. W., Donnelly, C. A., Fang, G.-R., & Webster, J. P. (2009). Evolution in a multi-host parasite: Chronobiological circadian rhythm and population genetics of *Schistosoma japonicum* cercariae indicates contrasting definitive host reservoirs by habitat. *International Journal for Parasitology*, *39*(14), 1581–1588. https://doi.org/10.1016/j.ijpara.2009.06.003

Lu, D.-B., Wang, T.-P., Rudge, J. W., Donnelly, C. A., Fang, G.-R., & Webster, J. P. (2010). Contrasting reservoirs for *Schistosoma japonicum* between marshland and hilly regions in Anhui, China—A two-year longitudinal parasitological survey. *Parasitology*, *137*(1), 99–110. https://doi.org/10.1017/S003118200999103X

Luo, F., Yang, W., Yin, M., Mo, X., Pang, Y., Sun, C., Zhu, B., Zhang, W., Yi, C., Li, Z., Wang, J., Xu, B., Feng, Z., Huang, Y., Lu, Y., & Hu, W. (2022). A chromosome-level genome of the human blood fluke *Schistosoma japonicum* identifies the genomic basis of host-switching. *Cell Reports*, *39*(1), 110638. https://doi.org/10.1016/j.celrep.2022.110638

Magdi, I., Botros, S., AA, M., William, S., Farghally, A., Tao, L. F., Day, T., & Bennett, J. L. (1999). Resistance to praziquantel: Direct evidence from *Schistosoma mansoni* isolated from Egyptian villagers. *The American Journal of Tropical Medicine and Hygiene*, *60*, 932–935. https://doi.org/10.4269/ajtmh.1999.60.932

McCullough, F. S. (1959). The susceptibility and resistance of *Bulinus* (*Physopsis) globosus* and *Bulinus (Bulinus) truncatus rohlfsi* to two strains of *Schistosoma haematobium* in Ghana. *Bulletin of the World Health Organization*, *20*(1), 75–85. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2537803/

Melman, S. D., Steinauer, M. L., Cunningham, C., Kubatko, L. S., Mwangi, I. N., Wynn, N. B., Mutuku, M. W., Karanja, D. M. S., Colley, D. G., Black, C. L., Secor, W. E., Mkoji, G. M., & Loker, E. S. (2009). Reduced susceptibility to praziquantel among naturally occurring Kenyan isolates of *Schistosoma mansoni*. *PLoS Neglected Tropical Diseases*, *3*(8), e504. https://doi.org/10.1371/journal.pntd.0000504

Minai, M., Hosaka, Y., & Ohta, N. (2003). Historical view of schistosomiasis japonica in Japan: Implementation and evaluation of disease-control strategies in Yamanashi Prefecture. *Parasitology International*, *52*(4), 321–326. https://doi.org/10.1016/S1383-5769(03)00047-3

Mintsa-Nguéma, R., Moné, H., Ibikounlé, M., Mengué-Ngou-Milama, K., Kombila, M., & Mouahid, G. (2014). Cercarial emergence pattern of *Schistosoma haematobium* from Libreville, Gabon. *Parasite*, *21*, 3. https://doi.org/10.1051/parasite/2014004

Moendeg, K. J., Angeles, J. M. M., Nakao, R., Leonardo, L. R., Fontanilla, I. K. C., Goto, Y., Kirinoki, M., Villacorte, E. A., Rivera, P. T., Inoue, N., Chigusa, Y., & Kawazu, S. (2017). Geographic strain differentiation of *Schistosoma*

japonicum in the Philippines using microsatellite markers. *PLOS Neglected Tropical Diseases*, *11*(7), e0005749. https://doi.org/10.1371/journal.pntd.0005749

Mouahid, G., Idris, M. A., Verneau, O., Théron, A., Shaban, M. M. A., & Moné, H. (2012). A new chronotype of *Schistosoma mansoni*: Adaptive significance. *Tropical Medicine & International Health: TM & IH*, *17*(6), 727–732. https://doi.org/10.1111/j.1365-3156.2012.02988.x

Mougeot, G., Golvan, Y. J., & Depernet, D. (1977). The nematode-trematode interaction in *Biomphalaria glabrata*, the vector of schistosomiasis mansoni in the West Indies and South America. *Bulletin de Societe Des Sciences Veterinaires et de Medecine Comparee de Lyon*, *79*(3), 151–156.

N'Goran, E., Brémond, P., Sellin, E., Sellin, B., & Théron, A. (1997). Intraspecific diversity of *Schistosoma haematobium* in West Africa: Chronobiology of cercarial emergence. *Acta Tropica*, *66*(1), 35–44. https://doi.org/10.1016/S0001-706X(97)00676-1

Nikolakis, Z. L., Adams, R. H., Wade, K. J., Lund, A. J., Carlton, E. J., Castoe, T. A., & Pollock, D. D. (2022). Prospects for genomic surveillance for selection in schistosome parasites. *Frontiers in Epidemiology*, *2*. https://www.frontiersin.org/articles/10.3389/fepid.2022.932021

Panitz, E. (1966). Evidence for Race Formation Among Human Schistosome Parasites. *Systematic Zoology*, *15*(2), 143–152. https://doi.org/10.2307/2411633

Parsons, D. A. J., Walker, A. J., Emery, A. M., Allan, F., Lu, D.-B., Webster, J. P., & Lawton, S. P. (2025). Evolution of antigenic diversity in the zoonotic multi-host parasite *Schistosoma japonicum*: Implications for vaccine design. *International Journal for Parasitology*, S0020-7519(25)00066-9. https://doi.org/10.1016/j.ijpara.2025.04.004

Parsons, D. A. J., Walker, A. J., Emery, A. M., Webster, J. P., & Lawton, S. P. (2023). Evolution of tetraspanin antigens in the zoonotic Asian blood fluke *Schistosoma japonicum. Parasites & Vectors*, *16*(1), 97. https://doi.org/10.1186/s13071-023-05706-3

Perrin, C., Lepesant, J. M. J., Roger, E., Duval, D., Fneich, S., Thuillier, V., Alliene, J.-F., Mitta, G., Grunau, C., & Cosseau, C. (2013). *Schistosoma mansoni* Mucin Gene (SmPoMuc) Expression: Epigenetic Control to Shape Adaptation to a New Host. *PLOS Pathogens*, *9*(8), e1003571. https://doi.org/10.1371/journal.ppat.1003571

Pinto-Almeida, A., Mendes, T., Armada, A., Belo, S., Carrilho, E., Viveiros, M., & Afonso, A. (2015). The Role of Efflux Pumps in S*chistosoma mansoni* Praziquantel Resistant Phenotype. *PLoS ONE*, *10*(10), e0140147. https://doi.org/10.1371/journal.pone.0140147

Pitchford, R. J., Meyling, A. H., Meyling, J., & Du Toit, J. F. (1969). Cercarial shedding patterns of various schistosome species under outdoor conditions in the Transvaal. *Annals of Tropical Medicine & Parasitology*, *63*(3), 359–371. https://doi.org/10.1080/00034983.1969.11686637

Platt II, R. N., Le Clec'h, W., Chevalier, F. D., McDew-White, M., LoVerde, P. T., Ramiro de Assis, R., Oliveira, G., Kinung'hi, S., Djirmay, A. G., Steinauer, M. L., Gouvras, A., Rabone, M., Allan, F., Webster, B. L., Webster, J. P., Emery, A. M., Rollinson, D., & Anderson, T. J. C. (2022). Genomic analysis of a parasite invasion: Colonization of the Americas by the blood fluke *Schistosoma mansoni*. *Molecular Ecology*, *31*(8), 2242–2263. https://doi.org/10.1111/mec.16395

Platt II, R. N., McDew-White, M., Le Clec'h, W., Chevalier, F. D., Allan, F., Emery, A. M., Garba, A., Hamidou, A. A., Ame, S. M., Webster, J. P., Rollinson, D., Webster, B. L., & Anderson, T. J. C. (2019). Ancient Hybridization and Adaptive Introgression of an Invadolysin Gene in Schistosome Parasites. *Molecular Biology and Evolution*, *36*(10), 2127–2142. https://doi.org/10.1093/molbev/msz154

Resch, A. M., Carmel, L., Mariño-Ramírez, L., Ogurtsov, A. Y., Shabalina, S. A., Rogozin, I. B., & Koonin, E. V. (2007). Widespread Positive Selection in Synonymous Sites of Mammalian Genes. *Molecular Biology and Evolution*, *24*(8), 1821–1831. https://doi.org/10.1093/molbev/msm100

Rey, O., Toulza, E., Chaparro, C., Allienne, J.-F., Kincaid-Smith, J., Mathieu-Begné, E., Allan, F., Rollinson, D., Webster, B. L., & Boissier, J. (2021a). Diverging patterns of introgression from *Schistosoma bovis* across *S. haematobium* African lineages. *PLOS Pathogens*, *17*(2), e1009313. https://doi.org/10.1371/journal.ppat.1009313

Rey, O., Webster, B. L., Huyse, T., Rollinson, D., Van den Broeck, F., Kincaid-Smith, J., Onyekwere, A., & Boissier, J. (2021). Population genetics of African *Schistosoma* species. *Infection, Genetics and Evolution*, *89*, 104727. https://doi.org/10.1016/j.meegid.2021.104727

Richards, L., Erko, B., Ponpetch, K., Ryan, S. J., & Liang, S. (2019). Assessing the nonhuman primate reservoir of *Schistosoma mansoni* in Africa: A systematic review. *Infectious Diseases of Poverty*, *8*, 32. https://doi.org/10.1186/s40249-019-0543-7

Roquis, D., Rognon, A., Chaparro, C., Boissier, J., Arancibia, N., Cosseau, C., Parrinello, H., & Grunau, C. (2016). Frequency and mitotic heritability of epimutations in *Schistosoma mansoni*. *Molecular Ecology*, *25*(8), 1741–1758. https://doi.org/10.1111/mec.13555

Rudge, J. W., Carabin, H., Balolong, E., Tallo, V., Shrivastava, J., Lu, D.-B., Basáñez, M.-G., Olveda, R., McGarvey, S. T., & Webster, J. P. (2008). Population Genetics of *Schistosoma japonicum* within the Philippines Suggest High

Levels of Transmission between Humans and Dogs. *PLoS Neglected Tropical Diseases*, *2*(11), e340. https://doi.org/10.1371/journal.pntd.0000340

Saijuntha, W., Tantrawatpan, C., Agatsuma, T., Duenngai, K., Sithithaworn, P., Petney, T. N., & Andrews, R. H. (2020). Intron sequence variation of the echinostomes (Trematoda; Echinostomatidae): Implications for genetic investigations of the 37 collar-spined, *Echinostoma miyagawai* Ischii, 1932 and *E. revolutum* (Fröelich, 1802). *Parasitology Research*, *119*(8), 2485–2494. https://doi.org/10.1007/s00436-020-06734-z

Saijuntha, W., Tantrawatpan, C., Agatsuma, T., Wang, C., Intapan, P. M., Maleewong, W., & Petney, T. N. (2018). Revealing genetic hybridization and DNA recombination of *Fasciola hepatica* and *Fasciola gigantica* in nuclear introns of the hybrid Fasciola flukes. *Molecular and Biochemical Parasitology*, *223*, 31–36. https://doi.org/10.1016/j.molbiopara.2018.06.004

Sanchez, M. C., Cupit, P. M., Bu, L., & Cunningham, C. (2019). Transcriptomic analysis of reduced sensitivity to praziquantel in *Schistosoma mansoni*. *Molecular and Biochemical Parasitology*, *228*, 6–15. https://doi.org/10.1016/j.molbiopara.2018.12.005

Su, J., Zhou, F., & Lu, D.-B. (2013). A circular analysis of chronobiology of *Schistosoma japonicum* cercarial emergence from hilly areas of Anhui, China. *Experimental Parasitology*, *135*(2), 421–425. https://doi.org/10.1016/j.exppara.2013.08.010

Summers, S., Bhattacharyya, T., Allan, F., Stothard, J. R., Edielu, A., Webster, B. L., Miles, M. A., & Bustinduy, A. L. (2022). A review of the genetic determinants of praziquantel resistance in *Schistosoma mansoni*: Is praziquantel and intestinal schistosomiasis a perfect match? *Frontiers in Tropical Diseases*, *3*. https://www.frontiersin.org/articles/10.3389/fitd.2022.933097

Sun, H.-Y., Zhang, J.-Y., Zhang, H.-X., Xu, Q., & Lu, D.-B. (2023). Genetic difference between two *Schistosoma japonicum* isolates with contrasting cercarial shedding patterns revealed by whole genome sequencing. *Parasite*, *30*, 59. https://doi.org/10.1051/parasite/2023061

Tameim, O., Abdu, K., Gaddal, A. E., & Jobin, W. (1985). Protection of Sudanese irrigation workers from schistosome infections by a shift to earlier working hours. *The Journal of Tropical Medicine and Hygiene*. https://www.semanticscholar.org/paper/Protection-of-Sudanese-irrigation-workers-from-by-a-Tameim-Abdu/2580b3c67fcc2aea188e10abded63afb00832358

Tantrawatpan, C., Maleewong, W., Thanchomnang, T., Pilap, W., Agatsuma, T., Andrews, R., Sithithaworn, P., & Saijuntha, W. (2023). Intron Regions as Genetic Markers for Population Genetic Investigations of *Opisthorchis viverrini* sensu lato and *Clonorchis sinensis. Animals*, *13*, 3200. https://doi.org/10.3390/ani13203200

Tantrawatpan, C., Tapdara, S., Agatsuma, T., Sanpool, O., Intapan, P. M., Maleewong, W., & Saijuntha, W. (2021). Genetic differentiation of Southeast Asian *Paragonimus* Braun, 1899 (Digenea: Paragonimidae) and genetic variation in the *Paragonimus heterotremus* complex examined by nuclear DNA sequences. *Infection, Genetics and Evolution: Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *90*, 104761. https://doi.org/10.1016/j.meegid.2021.104761

Taylor, M. G., James, E. R., Nelson, G. S., Bickle, Q., Dunne, D. W., Dobinson, A. R., Dargie, J. D., Berry, C. I., & Hussein, M. F. (1977). Modification of the pathogenicity of *Schistosoma mattheei* for sheep by passage of the parasite in hamsters. *Journal of Helminthology*, *51*(4), 337–345. https://doi.org/10.1017/s0022149x00007690

Théron, A. (1984). Early and Late Shedding Patterns of *Schistosoma mansoni* Cercariae: Ecological Significance in Transmission to Human and Murine Hosts. *The Journal of Parasitology*, *70*(5), 652–655. https://doi.org/10.2307/3281744

Théron, A., & Combes, C. (1988). Genetic analysis of cercarial emergence rhythms of *Schistosoma mansoni*. *Behavior Genetics*, *18*(2), 201–209. https://doi.org/10.1007/BF01067842

Vianney, T. J., Berger, D. J., Doyle, S. R., Sankaranarayanan, G., Serubanja, J., Nakawungu, P. K., Besigye, F., Sanya, R. E., Holroyd, N., Allan, F., Webb, E. L., Elliott, A. M., Berriman, M., & Cotton, J. A. (2022). Genome-wide analysis of *Schistosoma mansoni* reveals limited population structure and possible praziquantel drug selection pressure within Ugandan hot-spot communities. *PLOS Neglected Tropical Diseases*, *16*(8), e0010188. https://doi.org/10.1371/journal.pntd.0010188

Wang, T. P., Shrivastava, J., Johansen, M. V., Zhang, S. Q., Wang, F. F., & Webster, J. P. (2006). Does multiple hosts mean multiple parasites? Population genetic structure of *Schistosoma japonicum* between definitive host species. *International Journal for Parasitology*, *36*(12), 1317–1325. https://doi.org/10.1016/j.ijpara.2006.06.011

Webster, B. L., Webster, J. P., Gouvras, A. N., Garba, A., Lamine, M. S., Diaw, O. T., Seye, M. M., Tchuem Tchuenté, L.-A., Simoonga, C., Mubila, L., Mwanga, J. R., Lwambo, N. J. S., Kabatereine, N. B., Lange, C. N., Kariuki, C., Mkoji, G. M., Rollinson, D., & Stothard, J. R. (2013). DNA 'barcoding' of *Schistosoma mansoni* across sub-Saharan Africa supports substantial within locality diversity and geographical separation of genotypes. *Acta Tropica*, *128*(2), 250–260. https://doi.org/10.1016/j.actatropica.2012.08.009

Weedall, G. D., & Conway, D. J. (2010). Detecting signatures of balancing selection to identify targets of anti-parasite immunity. *Trends in Parasitology*, *26*(7), 363–369. https://doi.org/10.1016/j.pt.2010.04.002

WHO. (2023). *Schistosomiasis*. https://www.who.int/news-room/fact-sheets/detail/schistosomiasis

Yang, Y., Wang, X.-Y., Duan, C., Wang, Z.-J., Sheng, H.-Y., Xu, X.-L., Wang, W.-J., & Yang, J.-H. (2023). Clinicopathological characteristics and its association with digestive system tumors of 1111 patients with Schistosomiasis japonica. *Scientific Reports*, *13*(1), Article 1. https://doi.org/10.1038/s41598-023-42456-9

Yin, M., Liu, X., Xu, B., Huang, J., Zheng, Q., Yang, Z., Feng, Z., Han, Z.-G., & Hu, W. (2016). Genetic variation between *Schistosoma japonicum* lineages from lake and mountainous regions in China revealed by resequencing whole genomes. *Acta Tropica*, *161*, 79–85. https://doi.org/10.1016/j.actatropica.2016.05.008

Yin, M., Zheng, H.-X., Su, J., Feng, Z., McManus, D. P., Zhou, X.-N., Jin, L., & Hu, W. (2015). Co-dispersal of the blood fluke *Schistosoma japonicum* and *Homo sapiens* in the Neolithic Age. *Scientific Reports*, *5*(1), Article 1. https://doi.org/10.1038/srep18058

Young, N. D., Chan, K.-G., Korhonen, P. K., Min Chong, T., Ee, R., Mohandas, N., Koehler, A. V., Lim, Y.-L., Hofmann, A., Jex, A. R., Qian, B., Chilton, N. B., Gobert, G. N., McManus, D. P., Tan, P., Webster, B. L., Rollinson, D., & Gasser, R. B. (2015). Exploring molecular variation in *Schistosoma japonicum* in China. *Scientific Reports*, *5*(1), Article 1. https://doi.org/10.1038/srep17345

Zahed, N. Z. (1992). Compatibility of *Biomphalaria arabica* to a baboon-adapted strain of *Schistosoma mansoni* from Saudi Arabia. *Saudi Medical Journal*, *13*(5), 419–422. https://pesquisa.bvsalud.org/gim/resource/pt/emr-26394

Zhao, Q. P., Jiang, M. S., Littlewood, D. T. J., & Nie, P. (2010). Distinct Genetic Diversity of *Oncomelania hupensis*, Intermediate Host of Schistosoma japonicum in Mainland China as Revealed by ITS Sequences. *PLoS Neglected Tropical Diseases*, *4*(3), e611. https://doi.org/10.1371/journal.pntd.0000611

Zhou, A., Zhang, W., Ge, X., Liu, Q., Luo, F., Xu, S., Hu, W., & Lu, Y. (2024). Characterizing genetic variation on the Z chromosome in *Schistosoma japonicum* reveals host-parasite co-evolution. *Parasites & Vectors*, *17*(1), 207. https://doi.org/10.1186/s13071-024-06250-4