Interventions and strategies for urogenital schistosomiasis elimination in Zanzibar

---

Inauguraldissertation

zur Erlangung der Würde einer Doktorin der Philosophie

vorgelegt der

Philosophisch-Naturwissenschaftlichen Fakultät

der Universität Basel

Von

Lydia Trippler

Basel, 2024

Genehmigt von der Philosophisch-Naturwissenschaftlichen Fakultät
auf Antrag von

Erstbetreuerin: Dr. Stefanie Knopp

Zweitbetreuer: Dr. Peter Steinmann

Externer Experte: Prof. Dr. Dr. Sören Becker

Basel, den 19.09.2023

_____

Prof. Dr. Marcel Mayor
Dekan der Philosophisch-Naturwissenschaftlichen Fakultät

“A disease outbreak anywhere is a risk everywhere.”

Tom Frieden

TABLE OF CONTENTS

ACKNOWLEDGEMENTS I

SUMMARY IV

TABLE OF ABBREVIATIONS VIII

1. GENERAL INTRODUCTION 1

1.1. Urogenital schistosomiasis 1

1.1.1. Transmission of the causative agent 1

1.1.2. Diagnosis 2

1.1.3. Interventions 3

1.1.4. Epidemiology and public health relevance 5

1.2. The study site 7

1.3. The studies 8

1.3.1. The Zanzibar Elimination of Schistosomiasis Transmission study 8

1.3.2. The SchistoBreak study 9

1.4. Rationale for goal and objectives 10

2. RESEARCH GOAL AND OBJECTIVES 11

3. PUBLICATIONS 12

3.1. The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones 12

3.2. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies 14

3.3. Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study 36

3.4. GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation 52

3.5. Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: baseline cross-sectional survey findings before the onset of a 3-year intervention study ... 65

3.6. Test-Treat-Track-Test-Treat (5T) approach for *Schistosoma haematobium* elimination on Pemba Island, Tanzania ... 81

4. DISCUSSION ... 98

4.1. The impact of large-scale interventions on schistosomiasis prevalence ... 98

4.1.1. Treatment interventions ... 98

4.1.2. Snail control interventions ... 100

4.1.3. Behavior change interventions ... 101

4.2. Micro-mapping of infection and environmental risk factors ... 104

4.3. Targeted alternatives to large-scale interventions in low-prevalence settings ... 107

5. CONCLUSION ... 110

REFERENCES ... 113

APPENDICES ... 126

Appendix A: Publication "The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones" 126

Appendix B: Supplementary material from publication "Test-Treat-Track-Test-Treat (5T) approach for *Schistosoma haematobium* elimination on Pemba Island, Tanzania" ... 248

ACKNOWLEDGEMENTS

I remember how I used to say that I would only consider doing a PhD if I found a topic that I would be really passionate about. And then you, Steffi, appeared in my life and presented me with a topic that I could not say no to. Thank you for all you have done for me during these last years. You gave me this fantastic opportunity about a fascinating and urgent topic, and all this in my favorite country. You have been my supervisor, you have been my role model in research and leadership, my mentor, and my caretaker in the middle of Pemba's nowhere when I was sick with food poisoning and high fever. Thank you for celebrating my wins with me and letting me make mistakes to learn from them. Thank you for always being only one phone call away. Thank you for having been the best supervisor I could have thought of.

Thank you, Peter and Jürg, for accompanying me over the years, for your feedback and invaluable input. Jürg and Jenny, thanks for making my first big international presentation possible! Jan, I have learned so much about statistics and coding from you and I highly appreciate all the hours and help you supported me with in the last 3.5 years. Thank you for never hesitating to share codes with me. Thank you, Aurelio, for helping me very much with ODK! Over the last years, I have met inspiring scientists and I have learned so much from all of you: David, Russell, Bonnie, Fiona, Lynsey, Poppy, Sergi, Tom (thanks for the nightshifts and for teaching me so much about snails!) & many more. Thank you!

A big thanks goes to the teams in Unguja and Pemba. This thesis was only possible with your dedication and hard work. This thesis is as much my success as it is yours. Thank you for every laughter and for all you have taught me. Thank you, Said, Shaali, Fatma & Saleh, for opening the doors to your islands, for teaching me in science, for guiding me, and for all your support. A big thanks to the whole Unguja team – when I came to Zanzibar in 2019, you have taught me so much about schistosomiasis and you have introduced me to all your work. You have showed me your beautiful island and I am very grateful that I started my Zanzibar journey with you! My deepest gratitude goes to the whole team in Pemba. We have spent many months together and I would not miss any for the world. Mohammed, Sarah and Khamisi, you have been rocking the surveillance part of SchistoBreak from the beginning, you are organizational stars, and I am so grateful for all your efforts! Ulfat & Anisa, thank you for your dedication to the behavior team and for all your passion to bring the message to the children and communities. Amour, Zahor, Massoud, Adam & Msanif, you have walked kilometers for the snail team, you have taught me so much, and I am very thankful we know each other! Khamisi,

Rama, Hemed, every time I am entering the lab I am smiling because you have such a positive mindset. Thank you for all you are doing every day! Ali, Ahmedi, Sele & Mbarouk, you have driven with us to the most remote areas of Pemba and always brought us back safe. Thank you for every kilometer. Luluwa, Amina & Feisal, without you we would not have any data to analyze. Thank you for your dedication! Thank you, Shadya, for doing such a great job with organizing our trips. Thank you, Bihawa for being my Pemba mother. Thank you Husna, Suleiman, and everybody from PHL. You are all part of the reason that SchistoBreak has been working so smoothly. Every time I arrive in Tanzania, it feels like coming home. This comes down to the people who make this place so very special. Ahsanteni sana!

I would not be who I am if it would not have been for my parents. Thank you for being with me and guiding me for 30 years. I was brave enough to move to Tanzania for one year when I was only 18 because you encouraged me and told me I could do it, and this experience has shaped my life tremendously. Thank you, Papa, for translating my first paper into German so everyone could read it and understand what I am doing in my work! Thank you, Tobi and Firat, for going with me through the highs and lows in the last years. Thank you for doing crossword puzzles with me on the phone, visiting me several times in Basel, and planting trees with me! Thank you, Mario and Melanie, for always having an open door for me. Mario, you have been there in very special moments of my life, which I will never forget! Talita and Raphael, you gave me a room when I needed it. Tali, you have been there for me in one of the darkest hours of my life, and I am forever grateful. Thank you for driving to train stations for me. I know you would do it again, and I love you for it. A couple of months ago, Steffi said that the Tripplers are a great family, and I fully agree. I am who I am because you all and Oma, Oma, and Opa made me the person I am today.

Thanks to all my hometown friends who have been supporting me from near and far over the course of this PhD. Anny, Annka, Marie and Rahel, being with you always makes me feel home. Lisa, you are my moral compass. Thank you for always being only a phone call away. Nici, thank you for always believing in me. Carsten and Stefan, whenever we meet, it feels like we just talked yesterday. Lea & Günni, thank you for your constant friendship. Danni and Detlef, you gave me a special room in your family with Helen and Janek; you always made time for me, and you came to visit me in different places several times. Thank you. And thank you, Heni, for encouraging me to do this PhD and for everything else.

And then a big thanks goes out to all my Basel friends. More than three years ago, I came here not knowing anybody, and I feel blessed that I met all of you. You are making my time in Basel just amazing, and I would not want to miss any of you. Francesca, coming all the way to Pemba, going with me through the highs and lows. You are amazing! Lyndsay, the patience and kindness in person. Thank you for reading, suggesting, correcting and being you! Alexandra, my solid rock. Your friendship means a lot to me! Nandi, my wise friend, your thoughts and friendship are invaluable to me. Adriana & Max, special people, special times, special hikes – thank you! Olga, my office-sunshine, your optimism is infectious. Anais, thanks for your kindness and passion and always having open ears. Philipp, Tanzania times, good times. Thanks for all your coding help. Alexandros, thanks for all the energy you gave me – and the cookies. Fakih, thanks for translation help. Christoph, Anna, Andi, Janine, Vanessa, Dani, Lotz, Isaac, Myrta, Alexis, Tanja, Behzad, Martha, Imma, Sophie, Martin, Naomi, and many more – I am so glad to know you and for having you in my life. You have made my PhD time unforgettable. A special thanks goes to Rory, my flatmate and friend, my partner in worms, and my English-corrector. Thank you for endless evenings on the balcony discussing life, work, god, and the world; for my own thesis-writing playlist, for listening, reading, and correcting. Thank you for always being only a couple of stairs away.

The last thanks goes to Toni, my partner, my friend, my biggest supporter. You celebrate my wins with me and support me through the lows. You have my back in the difficult times. You listen endlessly to my presentations, and give feedback. You remind me of what I can achieve when I tend to forget it. You find solutions with me when I cannot see them. You come home with my favorite chocolate after a hard day. You make me laugh like nobody else. You are the most generous person I know. You are amazing. Thank you for all of it.

SUMMARY

Urogenital schistosomiasis is caused by *Schistosoma haematobium* and can lead to severe morbidity if untreated. The human-snail-human life cycle of *S. haematobium* can be interrupted with treatment, snail control and behavior change interventions. The Zanzibar islands, United Republic of Tanzania, have achieved low overall prevalence nowadays; thus, their historical interventions can guide other sub-Saharan African countries towards the same goal. At the same time, questions arise about whether large-scale treatment is still justified in areas with a very low prevalence. Novel strategies are now needed to map environmental factors and human infection, identify clusters, and address spatial heterogeneity of infection focally.

The current thesis addresses these questions in several ways. A systematic review of 100 years of schistosomiasis and snail-related research on Zanzibar was performed to identify interventions and their impact on the *S. haematobium* prevalence. The impact of seven years of mass drug administration (MDA) and of a 16-month treatment gap on *S. haematobium* prevalence was determined. Finally, as part of the SchistoBreak study (2020-2024) being implemented to develop novel strategies for the elimination of schistosomiasis, two cross-sectional surveys in schools and households utilized micro-mapping to assess the impact of test-treat-track-test-treat (5T) interventions as an alternative to MDA in low-prevalence areas.

A hundred years of interventions resulted in a low overall prevalence of <5% in 2020. After a 16-month gap of MDA, spatial heterogeneity of *S. haematobium* infection on the islands was observed and became more pronounced when the prevalence rebounded primarily in hotspot areas. A novel strategy for infection mapping within the SchistoBreak study demonstrated the feasibility of finding pre-randomized households in remote settings. After one year of 5T interventions in low-prevalence areas, no significant prevalence increase was revealed.

A combination of MDA, including treating adults and preschool-aged children, snail control, and behavior change measures are crucial to reducing the *S. haematobium* prevalence in hotspot areas. Environmentally friendly snail control and new intervention grounds for behavior change measures need to be explored. To compare the schistosomiasis prevalence across countries and to create a global prevalence map, micro-mapping guidelines by the World Health Organization are required. In low-prevalence settings, targeted interventions present alternatives to MDA; however, future studies need to assess the optimal interventions required to maintain or further reduce the prevalence towards interruption of transmission.

ZUSAMMENFASSUNG

Die urogenitale Schistosomiasis wird durch *Schistosoma haematobium* verursacht und kann unbehandelt zu schwerer Morbidität führen. Der Mensch-Schnecke-Mensch-Lebenszyklus von *S. haematobium* kann durch medizinische Behandlung, Schneckenkontrolle und Verhaltensänderungen unterbrochen werden. In Sansibar, Vereinigte Republik Tansania, ist die Gesamtprävalenz heute sehr niedrig, so dass die dortigen historischen Maßnahmen anderen sub-Sahara afrikanischen Ländern als Vorbild dienen können. Gleichzeitig stellt sich die Frage, ob eine groß angelegte Behandlung in Gebieten mit einer sehr niedrigen Prävalenz noch gerechtfertigt ist. Es sind nun neuartige Strategien erforderlich, um Umweltfaktoren und menschliche Infektionen zu erfassen, Cluster zu identifizieren und die räumliche Heterogenität der Infektion gezielt anzugehen.

Eine systematische Übersichtsarbeit über 100 Jahre Schistosomiasisforschung in Sansibar wurde durchgeführt, um Interventionen und ihre Auswirkungen auf die Prävalenz von *S. haematobium* zu ermitteln. Es wurde ermittelt, wie sich eine siebenjährige Massenbehandlung mit Medikamenten (MDA) und eine 16-monatige Behandlungspause auf die Prävalenz von *S. haematobium* auswirkten. Die SchistoBreak-Studie (2020-2024) wird durchgeführt, um neue Strategien zur Eliminierung der Schistosomiasis zu entwickeln. In zwei Querschnittserhebungen in Schulen und Haushalten wurde mit Hilfe von Kartierung die Wirkung von Test-Behandlung-Nachverfolgung-Test-Behandlung (5T) als Alternative zur MDA in Gebieten mit niedriger Prävalenz bewertet.

Hundert Jahre Interventionen führten zu einer niedrigen Gesamtprävalenz von <5 % im Jahr 2020. Es wurde eine räumliche Heterogenität der *S. haematobium*-Infektionen auf den Inseln beobachtet, die besonders deutlich wurde, als die Prävalenz nach einer 16-monatigen Unterbrechung der MDA vor allem in Hotspot-Gebieten wieder anstieg. Eine neuartige Strategie zur Infektionskartierung im Rahmen der SchistoBreak-Studie zeigte, dass es möglich ist, prärandomisierte Haushalte in abgelegenen Gebieten zu finden. Nach einem Jahr 5T-Interventionen in Gebieten mit niedriger Prävalenz wurde kein signifikanter Anstieg der Prävalenz festgestellt.

Eine Kombination aus MDA, einschließlich der Behandlung von Erwachsenen und Vorschulkindern, Schneckenkontrolle und Maßnahmen zur Verhaltensänderung sind von entscheidender Bedeutung, um die Prävalenz von *S. haematobium* in Hotspot-Gebieten zu

verringern. Umweltfreundliche Schneckenkontrolle und neue Interventionsorte für Verhaltensänderungen müssen erforscht werden. Um die Prävalenz länderübergreifend vergleichen zu können und eine globale Prävalenzkarte zu erstellen, ist eine Micro-Mapping-Richtlinie der Weltgesundheitsorganisation erforderlich. In Gebieten mit niedriger Prävalenz stellen gezielte Interventionen eine Alternative zur MDA dar; künftige Studien sind jedoch erforderlich, um die optimalen Interventionen zu finden, um die niedrige Prävalenz aufrechtzuerhalten oder weiter zu senken.

MUHTASARI

Kichocho kinachoenea kwa njia ya mkojo husababishwa na kimelea kinachoitwa *Schistosoma haematobium*, na kinaweza kusababisha hali mbaya ya mwili ikiwa hakijatibiwa. Mzunguko wa maisha wa *S. haematobium* katika mwili wa binadamu unaweza kuzuiwa na matibabu, udhibiti wa konokono na mabadiliko ya tabia. Visiwa vya Zanzibar, vilivyopo ndani ya Jamhuri ya Muungano ya Tanzania, kwa sasa wamefikia kiwango cha chini cha maambukizi; Kwa hivyo, udhibiti wao wa kihistoria unaweza kuongoza nchi zingine za Afrika kusini mwa jangwa la sahara kuelekea lengo moja. Wakati huo huo, mjadala unaibuka; ikiwa matibabu ya watu wengi bado yanafaa katika eneo lenye kiwango cha chini sana cha maambukizi. Kwa sasa, mikakati mipya inahitajika kutoa mwelekeao wa sababu za kimazingira na maambukizo ya wanadamu, kubaini maeneo ya maambukizi, na kushughulikia utofauti uliopo katika maeneo mahsusi yanayopatikana maambukizi hayo.

Tasnifu hii inajadili maswali haya katika njia tofauti tofauti. Mapitio ya kimfumo ya miaka 100 ya kichocho na utafiti unaohusiana na konokono kwa Zanzibar ulifanyika ili kubaini udhibiti na athari zake za kuenea kwa *S. haematobium*. Matokeo ya ugawaji wa dawa kwa watu wengi (MDA) wa miaka saba, ambao uliacha muda wa miezi 16 bila ya kutoa matibabu ya kuenea kwa *S. haematobium* ulifanyika. Mwisho, utafiti wa Schistobreak (2020-2024) ulifanyika ili kuweka mikakati mipya ya kutokomeza kichocho. Tafiti mbili zilifanyika katika maeneo ya shule na kaya, tafiti hizi zilitumia ramani ndogo ndogo kutathmini athari za udhibiti uliojulikana kwa jina la chunguza-tibu-tafuta-chunguza-tibu (5T) kama njia mbadala ya MDA katika maeneo yenye maambukizi ya kiwango cha chini.

Udhibiti wa imaka mia moja kwa ujumla ulipelekea kiwango cha chini cha kuenea kwa kichocho cha <5% mnamo 2020. Baada ya muda wa miezi 16 bila ya kutoa matibabu, utofauti

katika maeneo wa maambukizi ya *S. haematobium* visiwani ulionekana kuongezeka zaidi hasa katika maeneo sugu. Mkakati mpya wa utafutaji wa maambukizi wa utafiti wa Schistobreak ulionyesha uwezekano wa kupata nyumba zilizobaikika kabla kwenye maeneo ya vijijini. Baada ya mwaka mmoja wa udhibiti wa 5T katika maeneo yenye maambukizi ya kiwango cha chini, hakukuonekana ongezeko la kiwango kikubwa cha maambukizi.

Mchanganyiko wa MDA, pamoja na matibabu ya watu wazima na watoto wenye umri mdogo wa kabla yakwenda shule, udhibiti wa konokono na hatua za mabadiliko ya tabia ni muhimu katika kupunguza kuenea kwa *S. haematobium* katika maeneo sugu. Njia rafiki kwa mazingira za udhibiti wa konokono na njia mpya za udhibiti za mabadiliko ya tabia zinahitaji kuchunguzwa zaidi. Ili kuweza kulinganisha kwa kuenea kwa ugonjwa wa kichocho baina ya nchi mbali mbali kwa lengo la kutoa matokeo ya jumla ya ramani ya dunia ya kuenea kwa kichocho, miongozo ya ramani ndogo ndogo kutoka kwa Shirika la afya Duniani inahitajika. Katika maeneo yenye maambukizi ya kiwango cha chini, hatua mahsusi zilizopo zinawasilisha mbadala kwa MDA; Ingawje, tafiti za baadae zinahitaji kutathmini uthibiti bora unaohitajika kudumisha au kupunguza zaidi kuenea kwa *S. haematobium* katika kuelekea udhibiti kabisa wa maambukizi.

TABLE OF ABBREVIATIONS

FGS	Female genital schistosomiasis
MDA	Mass drug administration
MGS	Male genital schistosomiasis
MoH	Ministry of Health
NTD	Neglected tropical disease
PHL-IdC	Public Health Laboratory – Ivo de Carneri
Swiss TPH	Swiss Tropical and Public Health Institute
WASH	Water, sanitation, and hygiene
WHO	World Health Organization
ZEST	Zanzibar Elimination of Schistosomiasis Transmission

1. GENERAL INTRODUCTION

1.1. Urogenital schistosomiasis

1.1.1. Transmission of the causative agent

There are three primary species of schistosomes that infect humans: *Schistosoma haematobium*, *S. mansoni*, and *S. japonicum*. The former causes urogenital schistosomiasis, while the latter two cause intestinal schistosomiasis [1]. This thesis focuses on the Zanzibar archipelago, United Republic of Tanzania, where *S. haematobium* is the only causative agent present [2], and thus targets urogenital schistosomiasis.

Schistosoma haematobium is a trematode parasite transmitted through a human-snail-human life cycle (Figure 1) [3]. Infected individuals who urinate in freshwater bodies release *S. haematobium* eggs into the water, which can stay viable for up to seven days [4]. Once the eggs hatch, free-living ciliated miracidia are released into the water. Snails of the genus *Bulinus* are suitable intermediate hosts for *S. haematobium* [5]. If a *Bulinus* snail is present in a freshwater body, the miracidia can penetrate the tissue of the snail where *S. haematobium* asexually reproduces through maternal and juvenile sporocyst stages within four to six weeks [3]. After replication, tens of thousands of cercariae are released into the freshwater body, potentially infecting humans. Snail populations and cercarial density have been shown to have strong spatial and seasonal variation [4]. Upon human skin penetration, the cercariae lose their tail and travel as schistosomula through the vasculature for the next 4-6 weeks. The parasites migrate through the lungs to the liver until they eventually settle, usually in the urogenital system, primarily in the veins surrounding the bladder, where sexually mature female and male worms pair and mate [4, 6]. Once paired, the female and male worm stay together and live an average of three to ten years in the human body [1]. Following fertilization, the female worm starts to release eggs into circulation. The eggs can transverse the wall of the blood vessel and of the bladder and enter the bladder lumen, from where they may be excreted from the human body through urine [6]. The *S. haematobium* life cycle restarts when an infected individual urinates in a freshwater body where *Bulinus* snails are present.

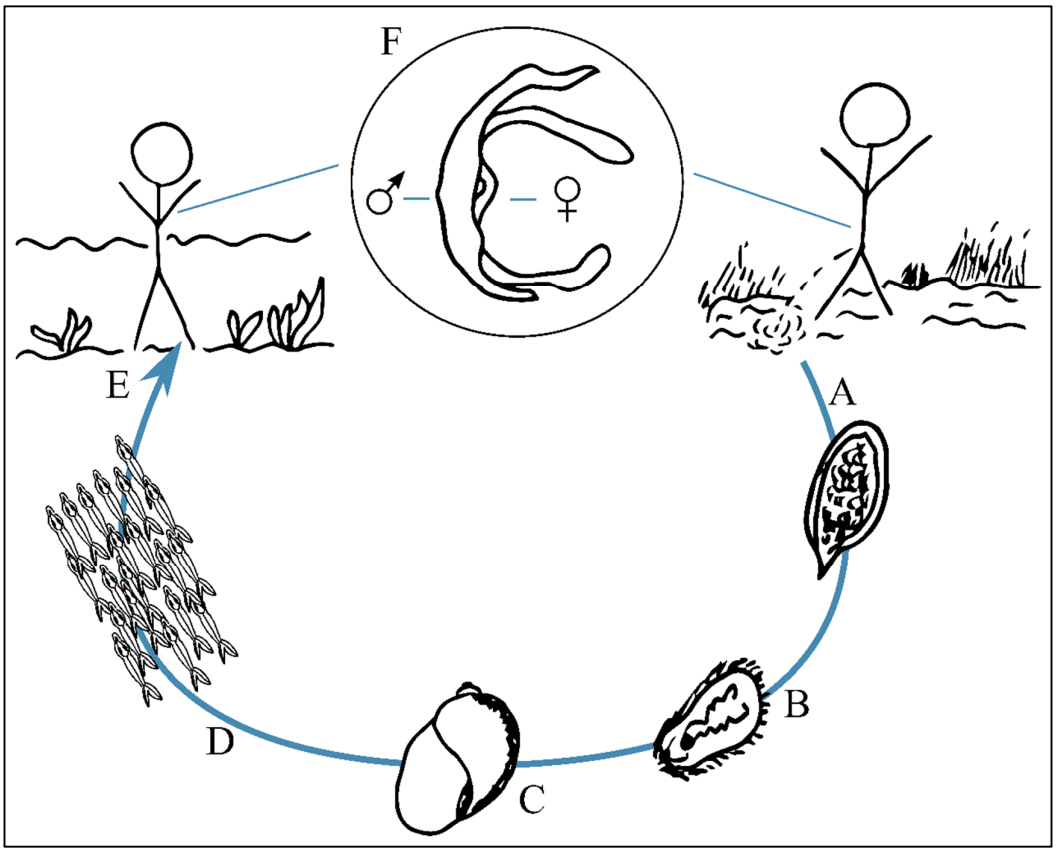


Figure 1. The life cycle of *Schistosoma haematobium*. A. An infected individual urinating into a freshwater body releases a *Schistosoma haematobium* egg; **B.** The egg hatches and releases a miracidium; **C.** The miracidium penetrates the tissue of the intermediate host snail of the *Bulinus Africanus* species group; **D.** Cercariae are released into the freshwater body; **E.** The cercariae penetrate the skin of another individual; **F.** The paired adult worms in the human body.

1.1.2. Diagnosis

The standard diagnostic method for detecting *S. haematobium* is to filter urine and look for ova under a light microscope [7]. The number of eggs indicates the intensity of the infection (light intensity: <50 eggs/10 ml urine; heavy intensity: ≥50 eggs / 10 ml urine); therefore, the standard amount of urine filtered is 10 ml [7, 8]. This urine filtration method shows a high sensitivity when used in high-prevalence areas, but the sensitivity of urine filtration decreases with decreasing overall prevalence in a region, and urine samples with ultra-low egg counts (<5 eggs/10 ml urine) are easily misinterpreted as false negatives [9]. The standard urine filtration method is not suitable for point-of-care testing as it requires a laboratory equipped with light microscopes. Mobile phone-based microscopes for testing *S. haematobium* infection have been developed, allowing point-of-care testing [10]. However, these microscopes show a low

sensitivity and may only serve as a viable alternative in resource-poor settings without a laboratory.

Another widely used method is to test for microhematuria (blood in the urine) as a proxy for *S. haematobium* infection [11-13]. Urine dipsticks, which change color when microhematuria is present, indicate the presence and intensity of blood in the urine. Studies have shown that the dipsticks are a good indicator of urogenital schistosomiasis in endemic settings [9, 14, 15]. However, as with urine filtration, the sensitivity of the dipsticks decreases in populations with low overall prevalence and in individuals with light-intensity infections [11, 15]. The specificity of microhematuria is lower than in urine filtration as microhematuria could also be the lead symptom for other health issues, such as genitourinary tract infections [16, 17]. Nonetheless, an advantage of testing for microhematuria is the ease of transportation and use of the dipstick allowing the test to be done at the point-of-care.

In addition to urine filtration and testing for microhematuria, other tests have been researched, such as antibody detection, antigen ELISA, circulating anodic antigen, recombinase polymerase amplification, and PCR assays [18-21]. Furthermore, there has been recent research on using clustered regularly interspaced short palindromic repeats (CRISPR) as a diagnostic tool for *S. haematobium* [22]. However, a low-cost test for urogenital schistosomiasis that is highly sensitive and specific, even in low-prevalence settings, and that can be used at the point of care is yet to be developed.

1.1.3. Interventions

The life cycle of *S. haematobium* (see 1.1.1.) can be interrupted at various stages, including killing the parasite in the human, killing the snails, and preventing individuals from urinating into freshwater bodies or coming into contact with water. In the absence of vaccines that could prevent humans from infection (and/or transmission) [23], the interventions that have been recommended by the World Health Organization (WHO) and have been implemented in various studies primarily focus on treatment, snail control, and behavior change [7, 24-26].

Preventive chemotherapy approaches were introduced for schistosomiasis and soil-transmitted helminths through inclusion in the World Health Assembly resolution 54.19 in 2001 [27]. Preventive chemotherapy with the drug of choice praziquantel against schistosomiasis has been implemented by many countries as mass drug administration (MDA) [28]. Praziquantel is administered at 40 mg/kg body weight in a single dose [29]. To simplify logistics in MDA, it is suggested to use a weight/height-adjusted dose pole as an alternative to bathroom-type scales

[30]. The safety of praziquantel has been established for individuals ≥4 years old [31]. Praziquantel shows high egg reduction rates in treated individuals but limited efficacy against juvenile worms, and it does not prevent reinfection [32, 33]. In 2001 with the World Health Assembly resolution 54.19, praziquantel MDA was recommended for school-age children as this age group is considered at the highest risk for infection and associated morbidity [4, 34]. As of 2022, the recommendation has been extended to preschool-aged children and adults, including pregnant women after the first trimester and lactating women [7]. Merck KGgA, the manufacturer of praziquantel, has pledged to donate praziquantel for an unlimited period for school-age children and, in certain regions, also for adults [35, 36]. In 2013, the question arose as to whether, and at what level of prevalence, MDA becomes unacceptable from a compliance, ethical, or cost perspective [37]. Since 2022, the WHO has recommended implementing treatment interventions on a community level rather than a country level based on the local prevalence of schistosomiasis [7]. In communities with ≥10% prevalence, (bi-)annual MDA is recommended, including all age groups from two years old in MDA programs with a coverage of ≥75% [7]. For communities with <10% prevalence, two different approaches may be implemented: i) continuing the formerly implemented MDA to move towards interruption of transmission or ii) implementing a test-and-treat approach targeting a specific population instead. The certainty of evidence for this recommendation is low, though, and additional research is needed to support and refine this recommendation.

To interrupt the life cycle at the snail level, snail control can be used to reduce the prevalence of schistosomiasis. Snail control is an umbrella term for interventions that include implementing environmental measures, such as stream channelization or drainage of irrigation schemes or using biological or chemical molluscicides [38]. Snail control aims to reduce the number of intermediate host snails in freshwater bodies by killing them or making the water bodies unattractive to snails. The WHO Pesticide Evaluation Scheme recommends the use of niclosamide, a chemical molluscicide, which has been specifically developed for the control of intermediate hosts for schistosomiasis and other trematodes, e.g., fascioliasis [38]. It is recommended to implement snail control activities in areas endemic for schistosomiasis in addition to human treatment, behavioral change, and water, sanitation, and hygiene (WASH) interventions [7]. However, niclosamide is also toxic to fish, invertebrates, and amphibians and should therefore be applied focally to minimize environmental damage [7].

The life cycle of *S. haematobium* can also be interrupted at the level of human behavior, as transmission requires human-waterbody contact [25]. Individuals are less likely to become

infected with *S. haematobium* if they can access alternatives to freshwater bodies for household chores or recreational activities [39]. In addition, individuals who do not urinate in freshwater bodies cannot spread *S. haematobium* and thus cannot sustain the life cycle [40]. Knowledge about the life cycle and the risks and pathogenesis of *S. haematobium* is vital to change human behavior [41]. Behavior change interventions, such as the encouragement to use toilet facilities, can help reduce the contact of individuals with freshwater bodies [25]. However, real behavior change can only come when alternatives to water bodies are provided, in particular WASH infrastructure [41]. Since 2022, the WHO has recommended implementing behavior change interventions in endemic areas alongside treatment, snail control, and WASH interventions [7].

1.1.4. Epidemiology and public health relevance

Schistosomiasis is a water-borne disease causing severe morbidity when untreated [4]. Schistosomiasis manifests in two phases, the acute and the chronic phase [1, 4]. Acute schistosomiasis occurs primarily in individuals infected for the first time, such as travelers, or may occur in individuals with severe reinfection [3]. Acute schistosomiasis occurs 14-84 days after infection and may include sudden onset of symptoms, such as fever, headache, eosinophilia, and fatigue [42].

Chronic schistosomiasis occurs primarily in individuals living in endemic areas [3]. Long-term morbidity caused by an infection with *S. haematobium* is due to an inflammatory response to eggs that do not complete the life cycle by exiting the human body through urination but instead become trapped in body tissues [43]. Severe urogenital schistosomiasis includes chronic urinary tract fibrosis (hydroureter and hydronephrosis) due to poor immune regulation of the human body's antischistosomal egg responses [1]. Untreated patients may also develop bladder cancer due to a urogenital schistosomiasis manifestation [44]. The leading symptom of urogenital schistosomiasis is blood in the urine, often accompanied by burning micturition and urinary frequency [1]. In addition, *S. haematobium* causes anemia due to blood loss and inflammation, and it impairs childhood development due to the effects of inflammation on physical fitness, cognitive function, iron metabolism, and expected growth [1]. Infection with *S. haematobium* can also cause morbidity related to the reproductive organs; female genital schistosomiasis (FGS) and male genital schistosomiasis (MGS) [1, 45]. Women with FGS may have sandy patches in the lower genital tract and lesions of the vagina, vulva, cervix, ovaries, and fallopian tubes [46]. FGS is associated with infertility and an increased risk of miscarriage [45]. In addition, there is growing evidence that FGS is associated with an increased risk of

sexually transmitted infections such as human immunodeficiency virus [47]. Pathology associated with MGS can be found in the prostate, testes, scrotum, and seminal vesicles. The leading symptom of MGS is hemospermia [48], but symptoms also include prostatitis, dyspareunia, oligospermia, and orchitis [1].

Despite its significant impact on the 251.4 million people at risk of infection with *Schistosoma* spp. in 78 countries worldwide, schistosomiasis belongs to the 20 neglected tropical diseases (NTDs) [7, 28, 36]. NTDs are defined as "*ancient diseases of poverty that impose a devastating human, social and economic burden on more than 1 billion people worldwide, predominantly in tropical and subtropical areas among the most vulnerable, marginalized population*" [36]. NTDs receive minimal attention within the global health agenda, resulting in limited resources and recognition by international funding agencies [49].

Schistosomiasis is found in various regions of the world, including The Americas, South-East Asia, and the Western Pacific, but it is mainly prevalent in Africa, where 90% of those requiring treatment live [28]. According to numbers from 2006, about 207 million people were infected worldwide with *Schistosoma* spp [50]. Sub-Saharan African countries are primarily affected by *S. mansoni* and *S. haematobium* [26]. Prevalence of *S. haematobium* in sub-Saharan Africa has been decreasing, with estimates of a population-adjusted prevalence of 17.4% in sub-Saharan Africa in 2010 to a prevalence of 6.2% in 2019 [26]. Typically, the highest infection prevalence and the highest prevalence of heavy-intensity infection are found in school-aged children, meaning that children are also primarily responsible for maintaining the transmission life cycle [4]. However, adults can also become infected, primarily those with frequent contact with freshwater bodies, e.g., for chores such as washing clothes/dishes, but also for bathing or fishing [1]. Infections in pre-school-age children are also increasingly recognized as important to combat [51].

1.2. The study site

The study site for all data and analyses presented in this thesis was the Zanzibar archipelago, consisting of the two islands Unguja and Pemba (Figure 2). The Zanzibar archipelago is a semi-autonomous part of the United Republic of Tanzania, and the islands are located around 30 km off the Tanzanian mainland. The islands are divided into 11 districts and subdivided into 129 small administrative areas known as "shehias" [52]. According to data collected through a census in 2022, the population of Unguja is about 1.3 million, with 900,000 people living in the urban area in western Unguja [53]. The population of Pemba is approximately 500,000, equally divided between northern and southern Pemba. In 2019/2020, 11.7% of the households on the Zanzibar islands did not have a toilet, particularly in the Micheweni district in northern Pemba, where 55.5% of the district population did not have a toilet [54]. In 2019/2020, only 57.0% of the households in Zanzibar had direct access to electricity, with a particularly low percentage in northern Pemba (15.7%).

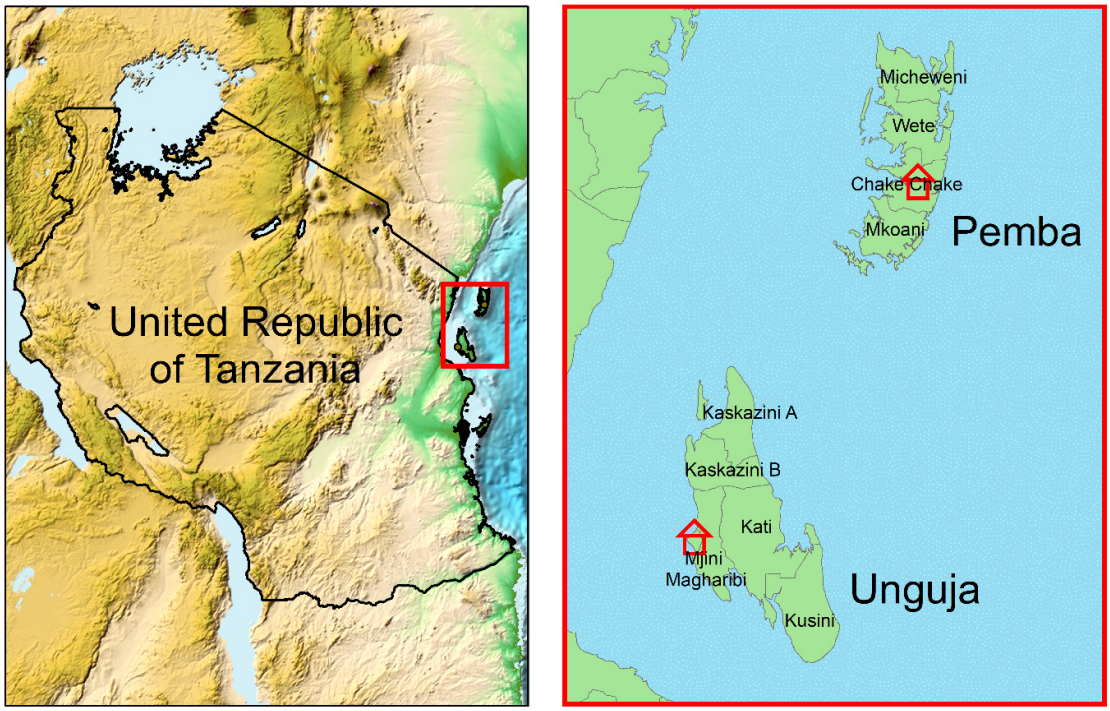


Figure 2. The United Republic of Tanzania (left) with the Zanzibar islands Unguja and Pemba (right) and the division of the islands into districts, and the location of the Neglected Tropical Diseases Center of the Zanzibar Ministry of Health on Unguja and the Public Health Laboratory – Ivo de Carneri on Pemba. The background of the left panel shows the bedrock topography of the United Republic of Tanzania and neighboring countries [55]. The district Mjini Magharibi on Unguja can be subdivided into the districts Mjini, Magharibi A, and Magharibi B.

All studies and results presented in this thesis were conducted and obtained with the support of Zanzibar authorities, primarily the NTD Program of the Zanzibar Ministry of Health (MoH) and the Public Health Laboratory – Ivo de Carneri (PHL-IdC).

On Unguja, the Zanzibar MoH's NTD Program has a laboratory in Lumumba, Mjini Magharibi (Figure 2), which is equipped to diagnose and monitor *S. haematobium* on the island and to implement control and elimination activities against *S. haematobium*, including mass drug administration, snail control, and behavior change interventions.

On Pemba, the PHL-IdC, constructed in 2000 in Wawi, Chake Chake, is a reference center for monitoring and evaluating national programs to control endemic diseases (Figure 2). The laboratory is well equipped for diagnosing and monitoring *S. haematobium* and other endemic diseases on Pemba and conducting control and elimination activities against *S. haematobium*, including MDA, snail control, and behavior change interventions. Since 2023, the PHL-IdC has also been equipped with a PCR laboratory.

1.3. The studies

The results of the presented PhD thesis are primarily based on two studies: the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) study and the SchistoBreak study.

1.3.1. The Zanzibar Elimination of Schistosomiasis Transmission study

In 2011, the ZEST study was formed by an alliance of the Neglected Tropical Diseases Program of the Zanzibar Ministry of Health, the PHL-IdC, the WHO, the Schistosomiasis Control Initiative (now Unlimit Health), the Natural History Museum London, the Swiss Tropical and Public Health Institute (Swiss TPH), and the Schistosomiasis Consortium for Operational Research and Evaluation (SCORE) [2, 56]. The ZEST study was designed as a large-scale cluster-randomized trial in 90 randomly selected shehias on Unguja and Pemba [56]. Each cluster consisted of 30 shehias (15 per island) and served as one out of three intervention arms: i) biannual MDA in schools and communities and snail control, ii) biannual MDA in schools and communities and behavior change interventions, and iii) biannual MDA in schools and communities alone. Parasitological surveys in schools and communities were conducted annually to monitor prevalence of *S. haematobium* on the islands and to assess the interventions' impact [57, 58]. (Bi-)annual MDA has continued to date, albeit with a 16-month

treatment gap in 2019/2020, following the completion of the study in 2017 [59]. The large-scale annual parasitological surveys in schools and communities were continued until 2021.

1.3.2. The SchistoBreak study

The SchistoBreak study is implemented in 20 shehias in the north of Pemba from 2020 to 2024 under the leadership of the Swiss TPH, the PHL-IdC, and the Zanzibar MoH. The study is designed as an intervention study to evaluate a multidisciplinary intervention approach to eliminate urogenital schistosomiasis in the north of Pemba [60]. In hotspot shehias, three complementary interventions are deployed: i) biannual MDA in schools and communities, ii) snail surveys in all known freshwater bodies and snail control if snails of the genus *Bulinus* are found, and iii) behavior change interventions in schools and communities. In low-prevalence shehias (<3% *S. haematobium* prevalence in schools and <2% *S. haematobium* prevalence in communities), no MDA is conducted. Instead, a surveillance-response approach is implemented, including active, passive, and reactive surveillance and reactive snail control. Annual parasitological surveys in schools and households are conducted to classify shehias as hotspot or low-prevalence implementation units and to monitor *S. haematobium* prevalence in all age groups of ≥4 years. Based on the parasitological surveys, the impact of the interventions in the north of Pemba is evaluated.

1.4. Rationale for goal and objectives

While schistosomiasis is endemic in 78 countries, 27 of those countries show low transmission [28]. The WHO defines the steps for countries to reach elimination of schistosomiasis into five sequential steps: i) morbidity control, ii) elimination as a public health problem, iii) interruption of transmission, iv) verification of elimination, and v) post-elimination surveillance [61]. One of the regions that shows a low transmission of *Schistosoma* infection and has achieved elimination as a public health problem (<1% heavy intensity infection prevalence) in 2017 is the Zanzibar archipelago [57].

After many years of various interventions to interrupt the life cycle of *S. haematobium*, such as treatment, snail control, and behavior change, overall prevalence of *S. haematobium* was assessed as 1.7% in 9-12 year old schoolchildren and as 1.5% in adults in 2017 [57, 58]. However, a spatial heterogeneity of *S. haematobium* infection on the islands has been observed down to the administrative level with many low-prevalence areas and a few hotspot areas [57]. Still, georeferenced data for Zanzibar allowing to implement targeted interventions are limited, as was long the case for large parts of Africa [37].

Much can be learned from the islands for future intervention implementation in Zanzibar and elsewhere by looking at historical interventions and their impact on the prevalence of *S. haematobium*, including the impact of the absence of interventions. Novel strategies are now needed to map *Schistosoma* infection in humans and environmental factors with geo-referenced data in order to identify and explain the infection clusters and to enable addressing the spatial heterogeneity of *S. haematobium* infection with focal interventions.

2. RESEARCH GOAL AND OBJECTIVES

The overarching goal of this PhD thesis is

Improving strategies and intervention approaches for schistosomiasis elimination in Zanzibar, United Republic of Tanzania.

To reach the goal, the following interrelated main objectives and specific research aims are pursued in the PhD thesis:

1. To document the interventions and the intervention's impact on the schistosomiasis prevalence in Zanzibar before 2021
    a. Identifying and discussing the milestones in schistosomiasis research, control, and elimination efforts from 1925 to 2022 on Unguja and Pemba, United Republic of Tanzania
    b. Monitoring *S. haematobium* prevalence in children and adults on Unguja and Pemba from 2012 to 2020 with a particular focus on the year 2020, after a 16-month treatment gap
2. To micro map *S. haematobium* infections and risk factors for infection in the north of Pemba from 2020-2024
    a. Presenting a study protocol for mapping and addressing the focality and heterogeneity of *S. haematobium* transmission, using novel adaptive intervention strategies for schistosomiasis elimination on Pemba
    b. Enhancing household-based randomized surveys for fine-scale mapping of *S. haematobium* prevalence in remote areas of Pemba, annually from 2020 to 2024
    c. Assessing risk factors for transmission and *S. haematobium* prevalence by mapping water bodies and human infections at the school- and household-level at the onset of the SchistoBreak study on Pemba in 2020/2021
3. To assess alternative interventions to mass drug administration against schistosomiasis in low-prevalence areas in the north of Pemba
    a. Assessing the perception of mass drug administration among the population in the north of Pemba and discussing focal interventions as alternatives
    b. Evaluating the performance of test-treat-track-test-treat interventions in implementation units with low prevalence of *S. haematobium* in the north of Pemba within the SchistoBreak study in 2021

3. PUBLICATIONS

3.1. The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones

Lydia Trippler[1,2], Stefanie Knopp[1,2], Sophie Welsche[3], Bonnie L. Webster[4,5], J. Russell Stothard[6], Lynsey Blair[3], Fiona Allan[4,5,7], Shaali Makame Ame[8], Saleh Juma[9], Fatma Kabole[8], Said Mohammed Ali[10], David Rollinson[4,5,11], Tom Pennance[4,5,12]

1 Swiss Tropical and Public Health Institute, Allschwil, Switzerland

2 University of Basel, Basel, Switzerland

3 Unlimit Health, London, United Kingdom

4 Natural History Museum, London, United Kingdom

5 London Centre for Neglected Tropical Disease Research, London, United Kingdom

6 Liverpool School of Tropical Medicine, Liverpool, United Kingdom

7 University of St Andrews, St Andrews, United Kingdom

8 Neglected Diseases Programme, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

9 Neglected Diseases Programme, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania

10 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

11 Global Schistosomiasis Alliance, London, United Kingdom

12 Western University of Health Sciences, Lebanon, Oregon, United States of America

This article was published in:

Advances in Parasitology (2023), Jul 17

doi: 10.1016/bs.apar.2023.06.001

Abstract

Zanzibar is among the few places in sub-Saharan Africa where interruption of *Schistosoma* transmission seems an achievable goal. Our systematic review identifies and discusses milestones in schistosomiasis research, control and elimination efforts in Zanzibar over the past 100 years. The search in online databases, libraries, and the World Health Organization Archives revealed 153 records published between May 1928 and August 2022. The content of records was summarised to highlight the pivotal work leading towards urogenital schistosomiasis elimination and remaining research gaps. The greatest achievement following 100 years of schistosomiasis interventions and research is undoubtedly the improved health of Zanzibaris, exemplified by the reduction in *Schistosoma haematobium* prevalence from > 50% historically down to < 5% in 2020, and the absence of severe morbidities. Experiences from Zanzibar have contributed to global schistosomiasis guidelines, whilst also revealing challenges that impede progression towards elimination. Challenges include: transmission heterogeneity requiring micro-targeting of interventions, post-treatment recrudescence of infections in transmission hotspots, biological complexity of intermediate host snails, emergence of livestock *Schistosoma* species complicating surveillance whilst creating the risk for interspecies hybridisation, insufficient diagnostics performance for light intensity infections and female genital schistosomiasis, and a lack of acceptable sanitary alternatives to freshwater bodies. Our analysis of the past revealed that much can be achieved in the future with practical implementation of integrated interventions, alongside operational research. With continuing national and international commitments, interruption of *S. haematobium* transmission across both islands is within reach by 2030, signposting the future demise of urogenital schistosomiasis across other parts of sub-Saharan Africa.

Due to the length of the article,
the complete publication can be found in the appendix of this thesis

3.2. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies

Lydia Trippler[1,2], Shaali Makame Ame[3], Jan Hattendorf[1,2], Saleh Juma[4], Salum Abubakar[4], Said Mohammed Ali[3], Fatma Kabole[4], David Rollinson[5], Stefanie Knopp[1,2]

[1] Swiss Tropical and Public Health Institute, Basel, Switzerland

[2] University of Basel, Basel, Switzerland

3 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

4 Neglected Diseases Programme, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

5 Natural History Museum, London, United Kingdom

This article was published in:

PLOS Neglected Tropical Diseases (2021), Feb 12; 15(2):e0009127

doi: 10.1371/journal.pntd.0009127

RESEARCH ARTICLE

Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: Repeated cross-sectional studies

Lydia Trippler[1,2], Shaali Makame Ame[3], Jan Hattendorf[1,2], Saleh Juma[4], Salum Abubakar[4], Said Mohammed Ali[3], Fatma Kabole[4], David Rollinson[5], Stefanie Knopp[1,2]*

1 Swiss Tropical and Public Health Institute, Basel, Switzerland, **2** University of Basel, Basel, Switzerland, **3** Public Health Laboratory—Ivo de Carneri, Pemba, United Republic of Tanzania, **4** Neglected Diseases Program, Ministry of Health, Zanzibar, United Republic of Tanzania, **5** Department of Life Sciences, Natural History Museum, London, United Kingdom

* s.knopp@swisstph.ch

OPEN ACCESS

Citation: Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. (2021) Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: Repeated cross-sectional studies. PLoS Negl Trop Dis 15(2): e0009127. https://doi.org/10.1371/journal.pntd.0009127

Editor: Antonio Montresor, World Health Organization, SWITZERLAND

Received: September 18, 2020

Accepted: January 12, 2021

Published: February 12, 2021

Data Availability Statement: The data that support the findings of this study from 2011/12-2017 are openly available in ClinEpiDB. The dataset "Study: SCORE Zanzibar *S. haematobium* Cluster Randomized Trial" can be found at https://clinepidb.org/ce/app/record/dataset/DS_eddb4757ba. The data that support the findings of this study from 2018-2020 are within the manuscript and its supporting information.

Abstract

Background

Considerable progress towards the elimination of urogenital schistosomiasis was made by the Zanzibar Elimination of Schistosomiasis Transmission project from 2012 till 2016, when biannual praziquantel mass drug administration (MDA) alone or with additional snail control or behaviour change interventions were implemented. Annual MDA was continued in 2017 and 2018, but not in 2019, imposing a 16-month treatment gap. We monitored the *Schistosoma haematobium* prevalence from 2012 till 2020 and assessed recrudescence patterns with focus on 2020.

Methodology

Repeated cross-sectional surveys were conducted from 2011/12 till 2020 in 90 communities and 90 schools in Zanzibar. Annually, around 4,500 adults and up to 20,000 schoolchildren were surveyed. The *S. haematobium* prevalence was detected by urine filtration and reagent strips. In 2020, risk factors for infection were investigated using generalized estimated equation models.

Principal findings

In adults, the apparent *S. haematobium* prevalence was 3.9% in 2011 and 0.4% in 2020. In schoolchildren, the prevalence decreased from 6.6% in 2012 to 1.2% in 2019 with vicissitudes over the years. Prominent recrudescence of infection from 2.8% in 2019 to 9.1% (+225%) in 2020 was observed in 29 schools with historically moderate prevalences (≥10%). Compared with 2019, reinfection in 2020 was particularly striking in boys aged

Funding: This study was funded by the University of Georgia Research Foundation Inc., which is funded by the Bill & Melinda Gates Foundation (https://www. gatesfoundation.org/) for the Schistosomiasis Consortium for Operational Research and Evaluation (SCORE; https://score.uga.edu/) projects (prime award no. 50816, sub-award no. RR374-053/4893206 to DR). SK received financial support by sub-award no. RR374-053/4893196 and via a direct grants from the Bill & Melinda Gates Foundation (Investment IDs: OPP1191423 and OPP1198086). Praziquantel was donated by the World Health Organization, Geneva, Switzerland and treatment implementation was supported by the Schistosomiasis Control Initiative, Imperial College, London, United Kingdom. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing interests: The authors have declared that no competing interests exist.

9–16 years. Being male was a risk factor for infection in 2020 (adults: odds ratio (OR): 6.24, 95% confidence interval (95% CI): 1.96–19.60; schoolchildren: OR: 2.06, 95% CI: 1.52–2.78). Living near to a natural freshwater body significantly increased the odds of infection in adults (OR: 2.90, CI: 1.12–7.54).

Conclusions/Significance

After 11 rounds of MDA over 7 years and a 16-month treatment gap, the urogenital schistosomiasis prevalence considerably rebounded in hotspot areas. Future elimination efforts in Zanzibar should focus on re-intensifying MDA plus additional interventions in hotspot areas. In low-prevalence areas, the strategy might be adapted from MDA to targeted surveillance-response.

Author summary

Schistosomiasis is a neglected tropical disease caused by parasitic blood flukes of the genus *Schistosoma*. On the Zanzibar islands, United Republic of Tanzania, interventions to eliminate urogenital schistosomiasis commenced in 2012. From 2012 to 2016, the population was treated biannually with praziquantel and, additionally, some areas received molluscicide against the intermediate host snail, or educational measures for behavior change. Mass drug administration (MDA) with praziquantel was continued annually in 2017 and 2018, but not in 2019. As a result of the interventions, the overall *S. haematobium* prevalence was reduced to 0.4% in adults and 3.4% in schoolchildren in 2020. However, in some areas, the MDA gap in 2019 resulted in a considerable rebound of infections. The recrudescence in 2020 was particularly striking for boys aged 9–16 years. In general, in 2020, male participants had higher odds of infection than females. Adults living near to a natural freshwater body also showed an increased risk of *S. haematobium* infection. Future elimination efforts in Zanzibar should focus on re-intensifying elimination interventions, including MDA, snail control and behavior change in hotspot areas. In low-prevalence areas, the strategy might be adapted from MDA to targeted interventions, such as surveillance-response.

Introduction

The neglected tropical disease (NTD) schistosomiasis is endemic in 78 countries worldwide and, according to estimates from 2017, it is responsible for about 1.4 million disability-adjusted life years annually [1,2]. In their new road map for NTDs 2021–2030, the World Health Organization (WHO) highlights the global elimination of schistosomiasis as a public health problem as target for 2030 [3]. Elimination as public health problem is therein defined as <1% proportion of heavy intensity infections [3]. According to mathematical modelling as well as multi-country and multi-year studies, elimination of schistosomiasis as public health problem can be achieved relatively quickly [4–6]. Once countries or areas have achieved this goal, they likely wish to move further down the elimination road, aiming for the interruption of schistosomiasis transmission, defined as zero incidence of infection.

Clearly, as long as interruption of transmission has not been achieved, recrudescence of transmission and disease remains a threat. Recrudescence of transmission can easily occur due

to the asexual reproduction of the parasite in its intermediate host snail and the release of thousands of cercariae into natural freshwater, which are then able to infect the next definitive human host [7,8]. Hence, as long as some, even very few people remain infected with schistosomes, the parasite's lifecycle can be perpetuated and the risk of recrudescence remains [9]. Also after the transmission of the parasite was successfully interrupted in an area or country, transmission can be reintroduced as long as the intermediate host snails thrive and *Schistosoma* eggs are released into the snails' freshwater habitat. To avoid recrudescence in areas where interruption of transmission has not yet been achieved, control and elimination efforts need to be maintained. To prevent reintroduction of schistosomiasis in areas considered post-elimination settings, a sensitive detection and timely treatment of cases through effective surveillance-response mechanisms is necessary [3]. Regarding the clonal reproduction of the parasite in its intermediate host snail, the detection of the parasite in the snail by xenomonitoring will constitute an important part of surveillance activities [7,8].

Interventions to control the morbidity caused by urogenital schistosomiasis in Zanzibar started in the 1980's when *S. haematobium* was highly prevalent on the islands Pemba and Unguja [10–13]. Control interventions were enforced by large-scale mass drug administration (MDA) campaigns in schools implemented in the early 2000s [14–16]. These interventions paved the way for one of the first projects to eliminate urogenital schistosomiasis from an area in sub-Sahara Africa: the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) project, implemented in Pemba and Unguja from 2011 till 2017 [17,18]. Over the course of ZEST, considerable progress towards the elimination of urogenital schistosomiasis was made [6,19]. During the ZEST project, MDA with praziquantel was implemented biannually in schools and whole communities across the islands Pemba and Unguja. In addition to biannual MDA, snail control and behavioural change interventions were implemented in 30 selected shehias (smallest administrative areas) in Pemba and Unguja, respectively, as part of a cluster randomized trial supported by the Schistosomiasis Consortium for Operational Research and Evaluation (SCORE) [17]. The impact of the interventions was monitored in annual cross-sectional surveys in 90 study schools and shehias from 2012 until 2017. The interventions reduced the *S. haematobium* prevalence in Zanzibar from 3.9% to 1.5% in 20–55 year old adults, and from 6.6% to 1.9% in 9–12 year old schoolchildren from 2012 to 2017. In 2017, heavy *S. haematobium* infection intensities ($\geq$50 eggs per 10 ml urine) occurred in 0.1% of 20–55 year old adults and in 0.5% of 9- to 12-year old schoolchildren [19]. Hence, within the ZEST project, urogenital schistosomiasis was eliminated as public health problem from most schools and communities in Zanzibar in 2017 and the *S. haematobium* prevalence was significantly reduced [6,19]. While the SCORE trial was concluded in early 2017, MDA in schools and shehia communities continued under the lead of the Zanzibar NTD Program, and was implemented annually in 2017 and 2018. No MDA was conducted in 2019 due to procurement issues.

We aimed to monitor the *S. haematobium* prevalence in children and adults in Zanzibar from 2012 till 2020 and to assess recrudescence patterns and risk factors for infection across all age groups with a particular focus on the year 2020, after the 16-month treatment gap.

Methods

Ethics statement

The SCORE cluster randomized intervention trial, including annual cross-sectional surveys to assess the *S. haematobium* prevalence in 90 study schools and shehias in Pemba and Unguja from 2012 till 2018 was approved by the Zanzibar Medical Research Ethics Committee in Zanzibar, United Republic of Tanzania (ZAMREC, reference no. ZAMREC 0003/Sept/011), the

Ethikkommission beider Basel (EKBB) in Basel, Switzerland (reference no. 236/22) and the Institutional Review Board of the University of Georgia in Athens, Georgia, United States of America (project no. 2012-10138-0). The trial is registered in the International Standard Randomized Controlled Trial Number register (ISRCTN; ISRCTN48837681). The cross-sectional surveys conducted in 2019 and 2020 obtained ethical approval from the Zanzibar Health Research Institute in Zanzibar (ZAHRI), United Republic of Tanzania (reference no. NO. ZAHREC/02/June/2019/36) and the Ethics Committee Northwest and Central Switzerland (EKNZ) in Basel, Switzerland (Project ID: Req-2019-00524). The surveys are registered in ISRCTN (ISRCTN17656730). All individuals participating in the cross-sectional surveys from 2011/12 until 2020 were informed in detail about the objectives and procedures of the studies and provided written informed consent for their participation. In the case of children below the age of 18, written consent was obtained from their parents or guardians.

Study site

The Zanzibar islands (Pemba and Unguja) are a semi-autonomous part of the United Republic of Tanzania. Both islands are located in the Indian Ocean, about 30 km off the coast from the east African mainland [20]. The islands are divided into 11 districts, which are again subdivided into 388 small administrative areas, called shehias [21]. The estimated population of Zanzibar for 2019 was 1.6 million with a population density of 669 person/km^2 in Unguja and 503 person/km^2 in Pemba. The proportion of the population aged 0–17 years was 49.0% [21]. Public primary schools in Zanzibar contain the grades 1–6 and some schools additionally include a nursery school. The net enrolment rate in primary schools was 83.2% in 2014/15 and the percentage of people being employed or full-time student was 67.8% [22]. Among the total population of Zanzibar, 28.9% had access to an improved water supply source in 2014/15 and 59.0% had access to an own improved toilet facility in 2015/16 [22,23].

Interventions for schistosomiasis elimination in Zanzibar

From 2012 to 2016, MDA was conducted biannually in Pemba and Unguja by the NTD Program of the Zanzibar Ministry of Health within the ZEST project [17]. The inhabitants of all shehias, excluding those located in the South district and in parts of the Urban district in Unguja, were offered praziquantel via community-wide treatment (CWT) or school-based treatment (SBT) approaches. During CWT, trained community drug distributors offered praziquantel (40 mg/kg) in a door-to-door approach to all community members aged >3 years that did not receive praziquantel in the same treatment round via SBT, or were not pregnant or severely sick, using a dose pole [24,25]. CWT was conducted biannually (twice a year) from April 2012 to November 2016 on both islands [6]. In SBT, trained teachers provided directly-observed praziquantel treatment to the children attending school on the day of treatment using a dose pole [24,25]. SBT was implemented biannually, from November 2013 until November 2016, with exception of 2014 where SBT was conducted only once a year in Pemba, but not at all in Unguja.

In addition to biannual MDA, 15 randomized shehias in Pemba and Unguja, respectively, received snail control interventions and 15 randomized shehias on each island received behaviour change interventions as part of the SCORE cluster randomized trial from 2012 till 2016. These interventions are described elsewhere in detail [6,17,19].

Once the SCORE trial and respective interventions were concluded in early 2017, the Zanzibar NTD Program continued the implementation of MDA. In 2017 and 2018, both CWT and SBT were conducted once a year. In 2019, no MDA was implemented, due to difficulties in procurement.

Study design and participants

The SCORE study implemented from 2011/12 until 2017 was designed as a 5-year cluster randomized trial. The impact of biannual MDA alone or in combination with snail control or behavior change interventions was monitored annually with cross-sectional surveys in a total of 90 shehias and 90 schools in Pemba and Unguja, respectively [17]. Details of the design of the SCORE study, including the justification of the number of participants, are described elsewhere [17,19]. In brief, the SCORE study population included annually ~50 adults aged 20- to 55 years in each of the 90 shehias that were enrolled in parasitological community-based surveys (CBS) that also included a questionnaire component. Moreover, ~100 schoolchildren aged 9- to 12-years from grade 3 and 4 in each of the 90 study schools were enrolled annually in parasitological school-based surveys (SBS) that also included a questioning component. Additionally, in the years 2012 and 2017, ~100 schoolchildren from grade 1 in each of the 90 study schools were enrolled in the SBS [19].

After the SCORE study was concluded in early 2017, the annual cross-sectional surveys continued with the same design and sample sizes for children aged 9- to 12-years in the SBS and for adults aged 20- to 55-years in the CBS in 2018, 2019, and 2020 to monitor the impact of annual MDA in 2017 and 2018 and the treatment gap in 2019 on *S. haematobium* infections.

In 2018, only a SBS including schoolchildren aged 9- to 12-years from grade 3 and 4 as well as children from grade 1 was conducted, but no CBS.

In 2019 and 2020, we were interested not only in the age groups targeted by SCORE, but aimed to investigate the extent of *S. haematobium* infections in a broader age group. Hence, in the CBSs in the years 2019 and 2020, not only 50 individuals aged 20–55 years as in SCORE, but 70 individuals aged 15–100 years were sampled per shehia. Moreover, in the SBS, the original sample size of ~100 children from grades 3 and 4 in each study school was kept as in SCORE, and additionally 20 children each from nursery, grade 1, 2, 5, and 6 were added.

The baseline CBS was conducted in November and December 2011. The baseline SBS was conducted from February till April 2012. All other CBS and SBS were conducted between January and May every year. Unfortunately, in 2020, the CBS and SBS were not completed due to the Covid-19 pandemic. Therefore, in 2020, the surveys were limited to 68 shehias and 67 corresponding schools (Fig 1).

Field procedures

Each year before the surveys commenced, the community leader (sheha) and head teacher of each study shehia and school, respectively, were invited to a meeting, where the procedures and aims of the forthcoming survey were explained and results from the previous survey were presented. The meeting participants were requested to inform their shehia population and school, respectively, about the purpose of the survey.

In the CBS, the survey teams visited each shehia for one day. In the years 2011–2017, data of ~50 adults per shehia were collected. In the years 2019 and 2020, data of ~70 adolescents and adults (aged 15–100 years) per shehia were collected. The randomization and selection of households and participants is described elsewhere in detail [17]. In all years, the study procedure was explained in Kiswahili to the members of the selected households by a trained fieldworker and the selected participants were asked to provide written consent by signing the informed consent form with their signature or thumbprint. Subsequently, they were invited to produce and submit a fresh urine sample and to answer questions from a pre-tested questionnaire. Besides the collection of demographic data, participants were asked about behaviors that might put them at risk of *S. haematobium* infection, for example their participation in the last MDA round, contact with natural freshwater, access to clean water, and travel history.

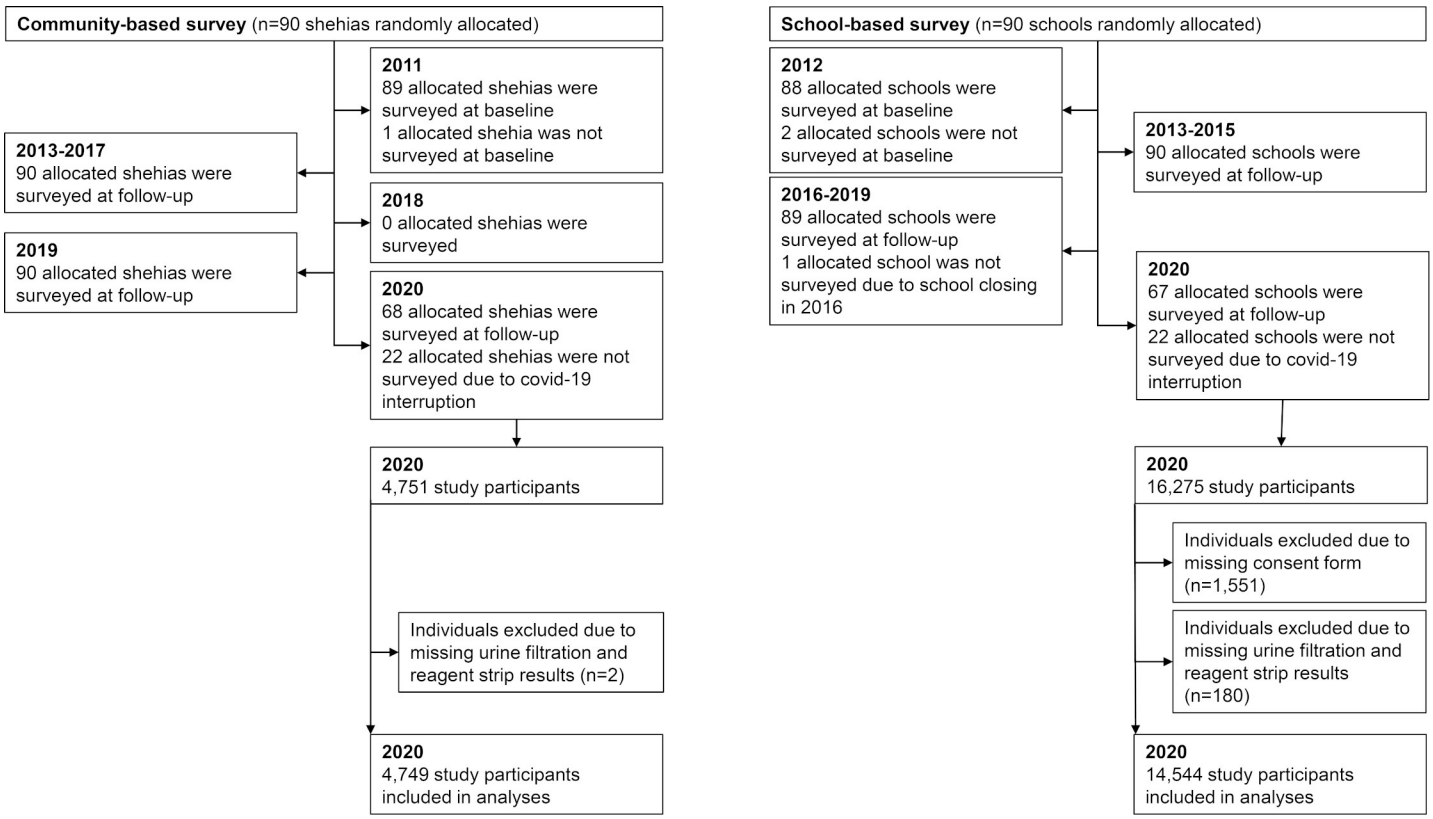


Fig 1. Flowchart of study design.

https://doi.org/10.1371/journal.pntd.0009127.g001

In the SBSs, each school was visited by the study team for two subsequent working days. The randomization of schoolchildren for participation is explained elsewhere in detail [17]. In brief, in each grade eligible for participation, girls and boys lined up stratified by sex, and every third child was selected until the requested number was reached. In 2012–2020, ~120 children from grades 3 and 4 were selected. In addition, in 2012, 2017 and 2018, ~120 children from grade 1 were selected. Finally, in 2019 and 2020, ~25 children each from grades 1, 2, 5 and 6 as well as from nursery were selected. The additional grades were included in 2019 and 2020 to obtain a better picture on the age profile of *S. haematobium* infections. The study procedure was explained in lay terms in Kiswahili to the enrolled children. Upon registration, the children were asked about their demographic data, their travel history, and in years with preceding MDA also about their participation in the last SBT round. All enrolled children received an information- and a consent-sheet for their parents or legal guardians to read and sign. On the next day, all children that submitted an informed consent form signed by their parents or legal guardians were given a plastic container (100 ml) to provide their fresh own urine sample.

All urine samples collected in CBS and SBS were produced between 10:00 and 14:00. The samples from Unguja were transferred to the laboratory of the NTD Program and all urine samples collected in Pemba were taken to the Public Health Laboratory–Ivo de Carneri (PHL-IdC).

Laboratory procedures

The urine samples were examined in the laboratories on the day of collection. First, the samples were investigated for visible macrohematuria using a color chart and for microhematuria

using reagent strips (Siemens Health Care AG, Zürich, Switzerland). Subsequently, each urine sample with a volume of at least 10 ml was vigorously shaken and filtered through a polycarbonate filter (Sefar Ltd., Bury, United Kingdom), which was then examined under a microscope for the presence of *S. haematobium* eggs by a well-experienced laboratory technician. The grade of macrohematuria and microhematuria, respectively, as well as the number of *S. haematobium* eggs for each participant was recorded on a laboratory form.

Outcomes

The primary outcome variable was the *S. haematobium* infection status in study participants from Pemba and Unguja in each cross-sectional survey.

The secondary analyses covered the annual *S. haematobium* prevalence and infection intensity as well as microhematuria levels among 20-to 55-year old adults and 9- to 12-year old children in the years 2011/12 to 2020 at aggregated and shehia/school level. Moreover, we assessed the age-prevalence distribution of *S. haematobium* infections in the years 2019 and 2020, stratified by sex, and including study participants of all ages. Finally, we determined the odds of infection in relation to demographic and behavioral factors in 2020.

Data management and statistical analyses

In all cross-sectional surveys from 2011/12 till 2020, data collected during registration in the SBS and laboratory examinations of the CBS and SBS were recorded on paper forms by the field enumerators and laboratory technicians, respectively, and subsequently double entered in electronic spreadsheets (Excel 2010, Microsoft) by data entry clerks in Zanzibar. In the years 2011/12 till 2019, paper questionnaire data collected in the CBS were entered into EpiInfo version 3.5.4 (Centers for Disease Control and Prevention, Atlanta, United States of America) by local staff in Zanzibar. In 2020, CBS questionnaire data were collected with the software Open Data Kit (https://opendatakit.org/) installed on Samsung Galaxy 4 tablets (Samsung Electronics, Seoul, South Korea). All data were cleaned and analyzed with StataIC 16 (StataCorp., Texas, United States of America) and R version 4.0.3 (www.rpoject.org).The data that support the findings of this study from 2011/12-2017 are openly available in ClinEpiDB. The dataset "Study: SCORE Zanzibar *S. haematobium* Cluster Randomized Trial" can be found at https://clinepidb.org/ce/app/record/dataset/DS_eddb4757ba. The data that support the findings of this study from 2018–2020 are within the manuscript and its supporting information (S1 Data and S1 Dictionary).

All study participants with a written informed consent and results of the urine filtration and/or reagent strip examination were included in the statistical analyses. A participant was considered *S. haematobium* positive if at least one egg per 10 ml urine was detected by the urine filtration method. Infections were stratified into light intensity (1–49 eggs per 10 ml urine) and heavy intensity ($\geq$50 eggs per 10 ml urine) infections, according to WHO guidelines [26]. In the absence of a urine filtration result, a participant was considered *S. haematobium*-positive if the urine was microhematuria-positive, as detected by reagent strips. The intensity of microhematuria was graded into negative, trace, 1, 2 or 3, in line with the color code provided by the reagent strip's manufacturer.

For the determination of the annual *S. haematobium* prevalence and infection intensity at aggregated and cluster level, only data from the 20-to 55-year old adults and from the 9-to 12-year old schoolchildren collected in the years 2011/12 to 2020 were included in the analyses. These restrictions were applied across the years, since during the SCORE study from 2011/12-2017, only data from these age groups were collected and we aimed to keep the annual data comparable.

For the differentiation of hotspot *versus* low prevalence areas, we defined a hotspot area as a shehia or school, which had a point prevalence of ≥10% in at least one of the study years and a low prevalence area as a shehia or school with point prevalences <10% throughout all study years from 2011/12 till 2020. Maps containing information about the spatial distribution of prevalence were created with R version 4.0.3. Coordinates of schools were collected with a handheld Garmin GPSMAP 62sc device (Garmin, Kansas City, USA). Shape files of shehias were provided by the Zanzibar Health Management Information System to the NTD Program of the Zanzibar Ministry of Health.

For the determination of the age profile of *S. haematobium* infections, participants of all ages of the 2019 and 2020 surveys were included in the analyses. Generalized estimating equation (GEE) models with exchangeable correlation structure were applied to estimate the odds ratios (OR) for *S. haematobium* infection. Separate models were run for the CBS and SBS, respectively, in 2020. The following "risk factors" were investigated as explanatory variables: sex (binary variable), age (categorical variable), travel history (binary variable), place of birth (categorical variable), living close to a natural freshwater body (binary variable; in CBS only), using water from natural freshwater bodies (binary variable; in CBS only), having a tap in or near the house (binary variable; in CBS only), having a well in or near the house (binary variable; in CBS only), and school grade (categorical variable; in SBS only). Besides the variables "place of birth" (excluded in both surveys due to collinearity), "use of water from natural freshwater bodies" (excluded in CBS due to collinearity), and "age" (excluded in SBS due to collinearity), all listed variables were included in the multivariable GEE models. In the models, 95% confidence intervals (CI) were used, and GEE with robust standard errors to account for clustering. Stratified by CBS and SBS, either the shehias (CBS) or the schools (SBS) were included in the model as clusters.

Results

Study flow and participant characteristics

The baseline CBS in 2011 was conducted in 89 among 90 allocated shehias in Pemba and Unguja; one allocated shehia was mistakenly not surveyed. In the CBSs implemented from 2013 to 2017 and in 2019, all 90 allocated shehias were surveyed. In 2020, the CBS was interrupted due to the Covid-19 pandemic, and only 68 among 90 allocated shehias were surveyed (Fig 1). In each of the CBS implemented from 2011 to 2017, around 4,400 adults aged 20–55 years participated. In the CBSs in 2019 and 2020, 6,300 and 4,749 individuals aged 15 years and older were enrolled.

The baseline SBS in 2012 was carried out in 88 among 90 allocated schools in Pemba and Unguja; two allocated schools were mistakenly not surveyed. In the SBSs implemented from 2013 to 2015, all 90 allocated schools were surveyed. Due to a school closing in 2016, the SBSs in 2016 till 2019 were conducted in 89 schools. Due to the Covid-19 pandemic and the associated interruption of survey activities in Zanzibar in March 2020, the SBS in 2020 was limited to 67 schools (Fig 1). In the SBSs conducted from 2012 to 2018, around 9,700 schoolchildren aged 9–12 years participated each year. In 2019 and 2020, a total of 19,559 and 14,544 children, respectively, were enrolled. Characteristics of the study participants from each CBS and SBS, respectively, are presented in Table 1.

Change in annual *S. haematobium* prevalence and intensity and microhematuria levels

Fig 2 shows that in the CBSs implemented from 2011 till 2020, the overall *S. haematobium* prevalence decreased constantly among the 20–55 year old adults from 3.9% in 2011 to 0.4% in 2020. An exception was 2014, when the prevalence increased by 0.5%-points (from 3.0% to

Table 1. Characteristics of the study participants from community-based survey (CBS) and school-based survey (SBS) by year.

Survey	Characteristic	Stratification	2011/12	2013	2014	2015	2016	2017	2018	2019	2020
CBS	**Shehias - n**		89	90	90	90	90	90	0	90	68
	Participants with outcome data - n		3974	4378	4462	4469	4474	4487	0	6300	4749
	Sex - n (%)	Female	2835 (71)	2671 (61)	2821 (63)	2478 (55)	2395 (54)	2133 (48)	0	3327 (53)	3010 (63)
		Male	1136 (29)	1707 (39)	1641 (37)	1991 (45)	2079 (46)	2354 (52)	0	2973 (47)	1739 (37)
	Age group - n	≤19 year old adults	0	0	0	0	0	0	0	1205	627
		20-55 year old adults	3974	4378	4462	4469	4474	4487	0	4742	3717
		≥56 year old adults	0	0	0	0	0	0	0	344	372
SBS	**Schools - n**		88	90	90	90	89	89	89	89	67
	Participants with outcome data - n		8931	9379	9595	9803	9725	10632	9662	19559	14544
	Sex - n (%)	Female	4664 (52)	5041 (54)	5098 (53)	5093 (52)	5122 (53)	5308 (50)	4903 (51)	9814 (50)	7407 (51)
		Male	4267 (48)	4338 (46)	4497 (47)	4710 (48)	4603 (47)	5324 (50)	4759 (49)	9745 (50)	7137 (49)
	Age group - n	≤8 year old children	0	0	0	0	0	0	0	4622	3606
		9-12 year old children	8931	9379	9595	9803	9725	10632	9662	12017	9081
		≥13 year old children	0	0	0	0	0	0	0	2906	1856

https://doi.org/10.1371/journal.pntd.0009127.t001

3.5%) compared with the previous year. The relative difference in the prevalence from 2011 to 2020 was -91%. Heavy intensity infections were found in 0.3% of participants at baseline in 2011, reached a maximum of 0.5% in 2014, and declined to zero in 2020.

Microhematuria levels in 20–55 year old adults were higher than the *S. haematobium* prevalence in all study years. From 2011 till 2020, the overall microhematuria among the 20–55 year old adults decreased from 12.2% in 2011 to 6.3% in 2020. A higher overall microhematuria level than in 2011 was observed in 2014 (15.3%). The highest grading of microhematuria (3) ranged between 1.0% and 3.7% and no decreasing trend in heavy microhematuria was observed over the study period.

In the SBS, a decreasing trend of the overall *S. haematobium* prevalence among the 9–12 year old children was observed from 2012 (6.6%) to 2019 (1.2%), with some inconsistent upturns in certain years (Fig 2). The relative difference in the prevalence from 2012 to 2019 was -82%. At baseline in 2012, heavy intensity infections were detected in 1.8% of participants and decreased to 0.3% in 2019 (-83%). A prominent rebound in the overall *S. haematobium* infections and intensity was observed after the 16-month treatment gap in 2020, when the prevalence of infections rose back from 1.2% to 3.4% (relative difference +183%) and heavy intensity infections rebounded from 0.3% to 0.8% (relative difference +167%).

Microhematuria levels in 9–12 year old schoolchildren were higher than the *S. haematobium* prevalence in all study years, except in 2013. In line with the *S. haematobium* prevalence, a decreasing trend of the overall microhematuria levels was observed from 2012 (9.5%) till 2019 (4.0%). The relative difference in the percentage of the overall microhematuria from 2012 to 2019 was -58%. At baseline in 2012, heavy microhematuria levels (3) were detected in 2.6% of participants and decreased to 1.2% in 2019 (-54%). A rebound in the overall microhematuria level was observed after the 16-month treatment gap in 2020, when the overall levels rose back from 4.0% to 5.2% (relative difference +30%) and heavy microhematuria rebounded from 1.2% to 2.5% (relative difference +108%).

Spatial heterogeneity of *S. haematobium* prevalence

The geographical distribution of the shehias and schools, and the *S. haematobium* prevalence in 20–55 year-old adults and 9–12 year-old children from 2011/12 to 2020 is indicated in Fig 3.

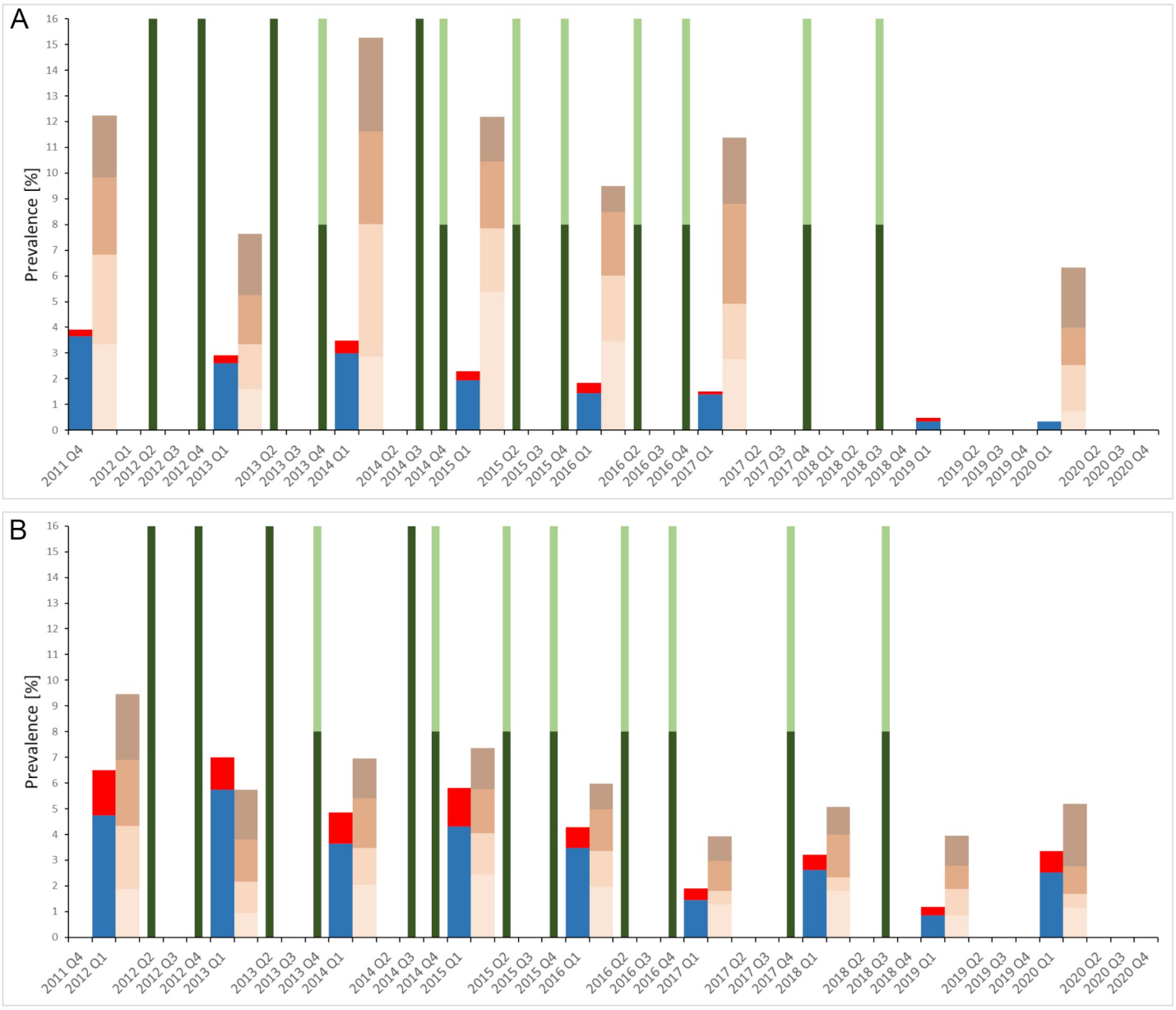


Fig 2. Prevalence and intensity of *S. haematobium* infections, microhematuria levels, and implementation of MDAs in Zanzibar from 2011/12 until 2020. The data show the *S. haematobium* prevalence and infection intensity, the levels of microhematuria, and implementation of MDA per annual quartile (Q). A: 20- to 55-year old adults; B: 9- to 12-year old schoolchildren. Blue: light intensity infection prevalence; red: heavy intensity infection prevalence. Very light brown: microhematuria graded trace; light brown: microhematuria graded 1; brown: microhematuria graded 2; dark brown: microhematuria graded 3. Dark green: community-wide treatment (CWT) only; dark and light green: community-wide (CWT) and school-based treatment (SBT).

https://doi.org/10.1371/journal.pntd.0009127.g002

A total of 71 shehias and 61 schools showed a prevalence <10% throughout all study years and were therefore defined as low prevalence areas, whereas 19 shehias and 29 schools crossed the 10% line in one or several years and were therefore considered hotspot areas. Fig 4 shows that the hotspot areas were more prone to recrudescence of *S. haematobium* infections and heavy intensity infections in one year or the other, and mainly responsible for the strong rebound observed after the 16-month treatment gap in 2020. While the rebound of the overall *S. haematobium* prevalence in schools from 2019 to 2020 was +183% (from 1.2% to 3.4%), the

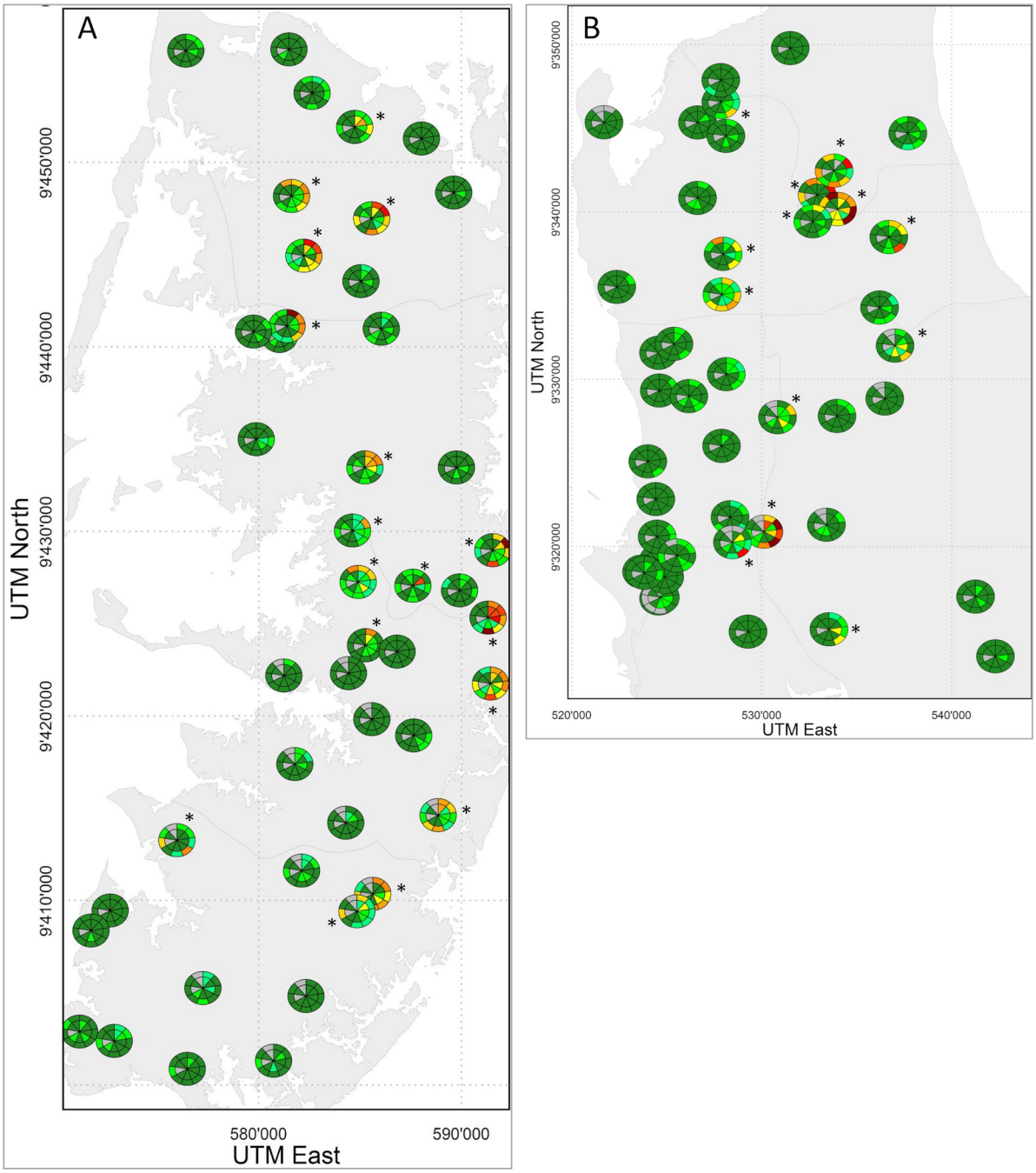


Fig 3. Maps indicating prevalence of *S. haematobium* infections from 2011/12 till 2020 in spatial clusters in Pemba (A) and Unguja (B). Data are *S. haematobium* prevalence in 45 schools/shehias (clusters) per island, stratified by population group and island. A: Pemba; B: Unguja. Data are presented per year starting from 2011/12 (12 o'clock position) to 2020. Inner circle: 20- to 55-year old adults; outer circle: 9- to 12-year old schoolchildren. The colors show the prevalence from dark green as lowest prevalence to dark red as highest prevalence, and grey indicates missing. The * represents a hotspot school and/or community. Maps containing information about the spatial distribution of prevalence were created with R version 4.0.3. Coordinates of schools were collected with a handheld Garmin GPSMAP 62sc device (Garmin, Kansas City, USA). Shape files of shehias were provided by the Zanzibar Health Management Information System to the Neglected Diseases Program of the Zanzibar Ministry of Health.

https://doi.org/10.1371/journal.pntd.0009127.g003

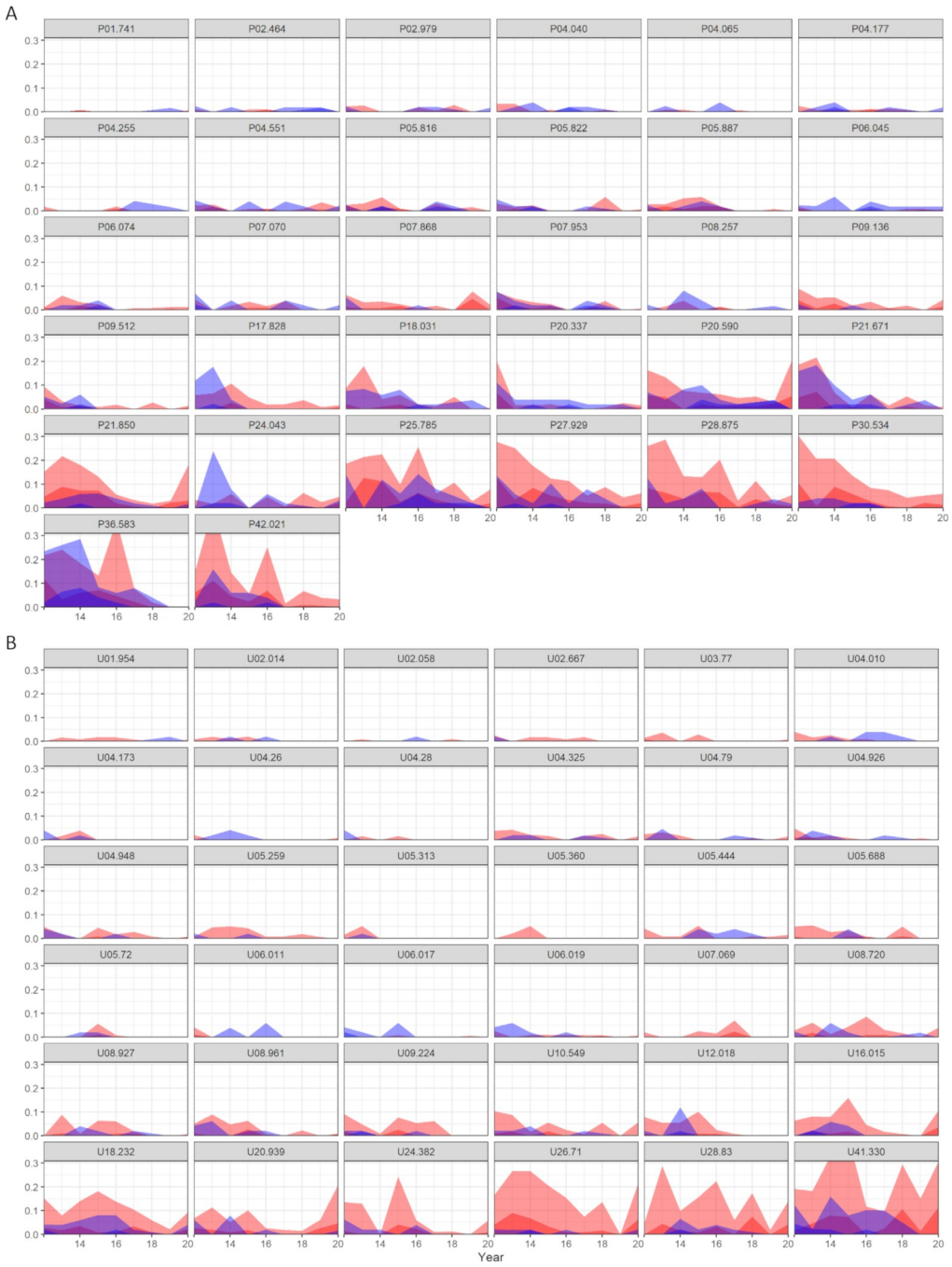


Fig 4. Prevalence of *S. haematobium* infections stratified by infection intensity levels in Zanzibar from 2011/12 until 2020 per spatial cluster in Pemba (A) and Unguja (B). Data are proportion of *S. haematobium* infection intensity in schoolchildren and adults in each surveyed cluster on each island. A: Shehias and schools in Pemba; B: Shehias and schools in Unguja. X-axis: year of survey (2011/12 to 2020); y-axis: proportion infection intensity. Dark blue: heavy intensity infection among 20- to 55-year old adults; light blue: light intensity infection among 20- to 55-year old adults. Dark red: heavy intensity infection among 9- to 12-year old schoolchildren; light red: light intensity infection among 9- to 12-year old schoolchildren.

https://doi.org/10.1371/journal.pntd.0009127.g004

rebound in hotspot areas was +225% (from 2.8% to 9.1%) and the rebound in the low prevalence areas was +75% (from 0.4% to 0.7%).

Age prevalence distribution

Fig 5 shows the age prevalence distribution of *S. haematobium* infections stratified by sex and in relation to the sampling effort in 2019 (Fig 5A) and 2020 (Fig 5B). In 2019, the *S. haematobium* infection levels were below 2.0% in both, male and female students, and community members. The prevalence was significantly higher in boys aged 8 to 16 years, compared with female students of the same age. No participant older than 42 years was tested *S. haematobium*-positive. In 2020, after the 16-month treatment gap, a prominent increase in prevalence was observed in 9–16 year-old schoolchildren of both sexes. Among the whole study population and all ages younger than 60 years, male participants had higher levels of infections than female participants did.

Risk factors for *S. haematobium* infection in 2020

In the CBS conducted in 2020, men had significantly higher chances of harboring a *S. haematobium* infection compared with women (OR: 6.24, 95% CI: 1.96–19.60, p = 0.002) (Fig 6A). Adults aged 20–55 years had significantly lower odds of having a *S. haematobium* infection than community members aged below 20 years (OR: 0.30, 95% CI: 0.13–0.69, p = 0.004). A *S. haematobium* infection was significantly associated with living in close proximity to a natural freshwater body (OR: 2.90, 95% CI: 1.12–7.54, p = 0.029). Having a tap in or near the house, travel outside the home island over the past 6 months, and praziquantel treatment within the last two years resulted in non-significant lower odds of *S. haematobium* infection.

In the SBS, boys had significantly higher chances of having a *S. haematobium* infection compared with girls (OR: 2.06, 95% CI: 1.52–2.78, $p < 0.001$) (Fig 6B). In each of the school grades 1 to 6, children had a higher chance of harboring a *S. haematobium* infection compared with children who attended nursery school, with children from grade 1 showing the lowest odds ratio (OR: 8.49, 95% CI: 1.89–38.16, p = 0.005) and children from grade 4 showing the highest odds ratio (OR: 15.70, 95% CI: 4.30–57.35, $p < 0.001$). A recent travel history outside the home island resulted in non-significantly lower odds of *S. haematobium* infection.

Discussion

Considerable progress towards the elimination of urogenital schistosomiasis was made by the ZEST project from 2012 till 2017, when biannual praziquantel MDA alone or with additional snail control or behaviour change interventions were applied. Annual MDA was continued across Zanzibar in 2017 and 2018, but not in 2019. We monitored the *S. haematobium* prevalence and microhematuria levels from 2012 until 2020 and assessed recrudescence patterns and risk factors for infection across all age groups with a particular focus on the year 2020, after the 16-month treatment gap.

We found that the apparent overall *S. haematobium* prevalence among adults decreased from 3.9% at baseline in 2011 to 0.35% in 2020. Overall microhematuria levels in adults decreased from 12.2% in 2011 to 6.3% in 2020. Microhematuria levels in adults were remarkably higher than the *S. haematobium* prevalence measured by egg output by urine filtration microscopy and also than the microhematuria levels in children. This observation might indicate that either many adults suffer from chronic morbidity due to urogenital schistosomiasis that is not reflected by the number of eggs excreted in urine, or, microhematuria in adults might be caused by reasons unrelated to schistosomiasis such as urinary tract infections, bladder stones, sickle cell anemia, or pregnancy or menstrual bleeding in females [27–29].

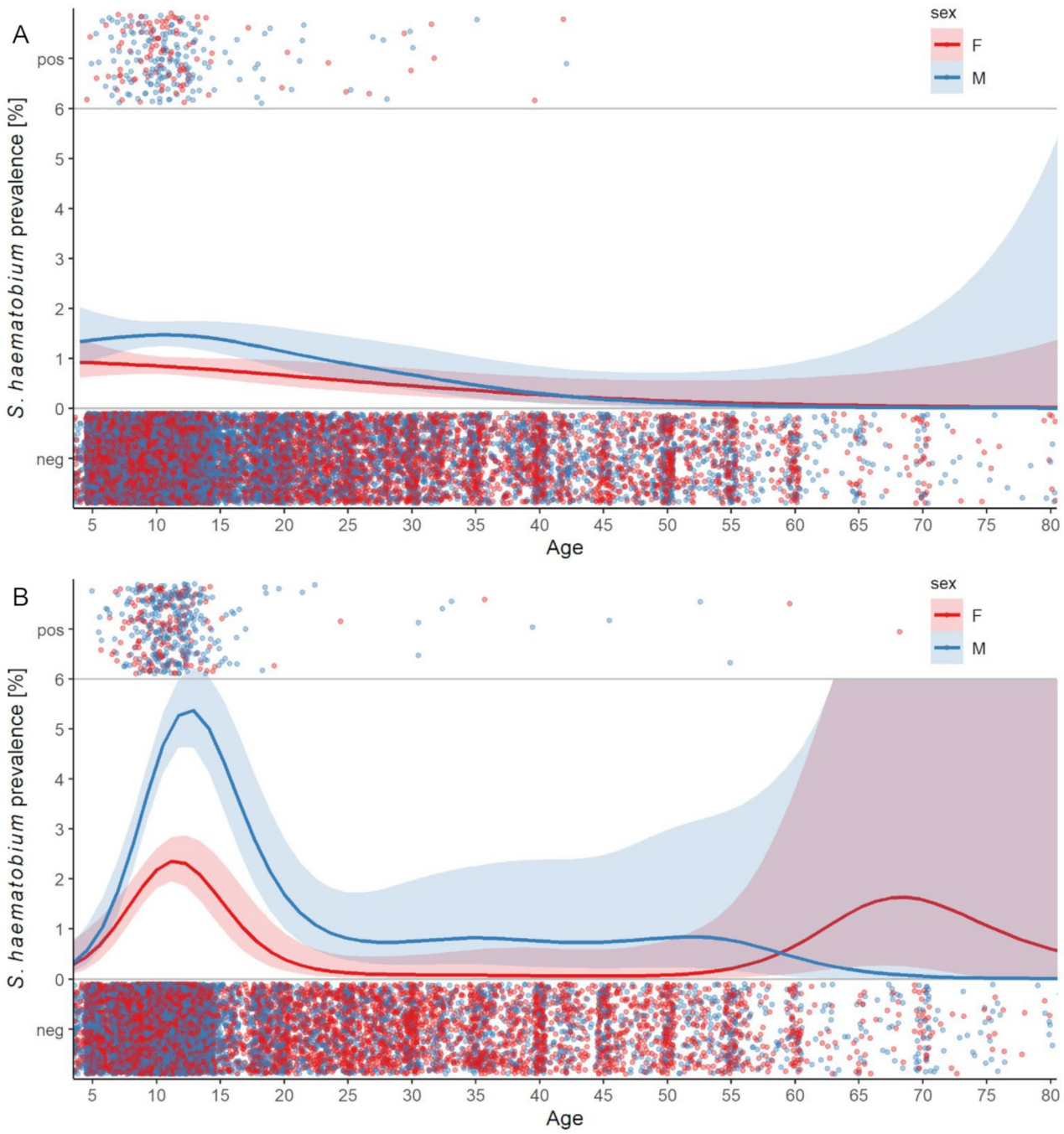


Fig 5. *S. haematobium* prevalence across all ages, stratified by sex, after 11 rounds of MDA in 2019 (A) and a 16-month treatment gap in 2020 (B). The blue (male) and red (female) dots in the upper part (pos) of each figure, each represent a *S. haematobium*-positive urine sample from an individual of a certain age (x-axis). The blue (male) and red (female) dots in the lower part (neg) of each figure, each represent a *S. haematobium*-negative urine sample prom an individual of a certain age (x-axis). The blue (male) and red (female) lines in the middle part of each figure represent the predicted *S. haematobium* age-prevalence. The shading around the lines represent 95% confidence bands.

https://doi.org/10.1371/journal.pntd.0009127.g005

The apparent overall *S. haematobium* prevalence among schoolchildren decreased from 6.6% at baseline in 2012 to 3.4% in 2020 and microhematuria levels from 9.5% to 5.2%, respectively. However, an even lower *S. haematobium* prevalence of 1.2% and microhematuria levels

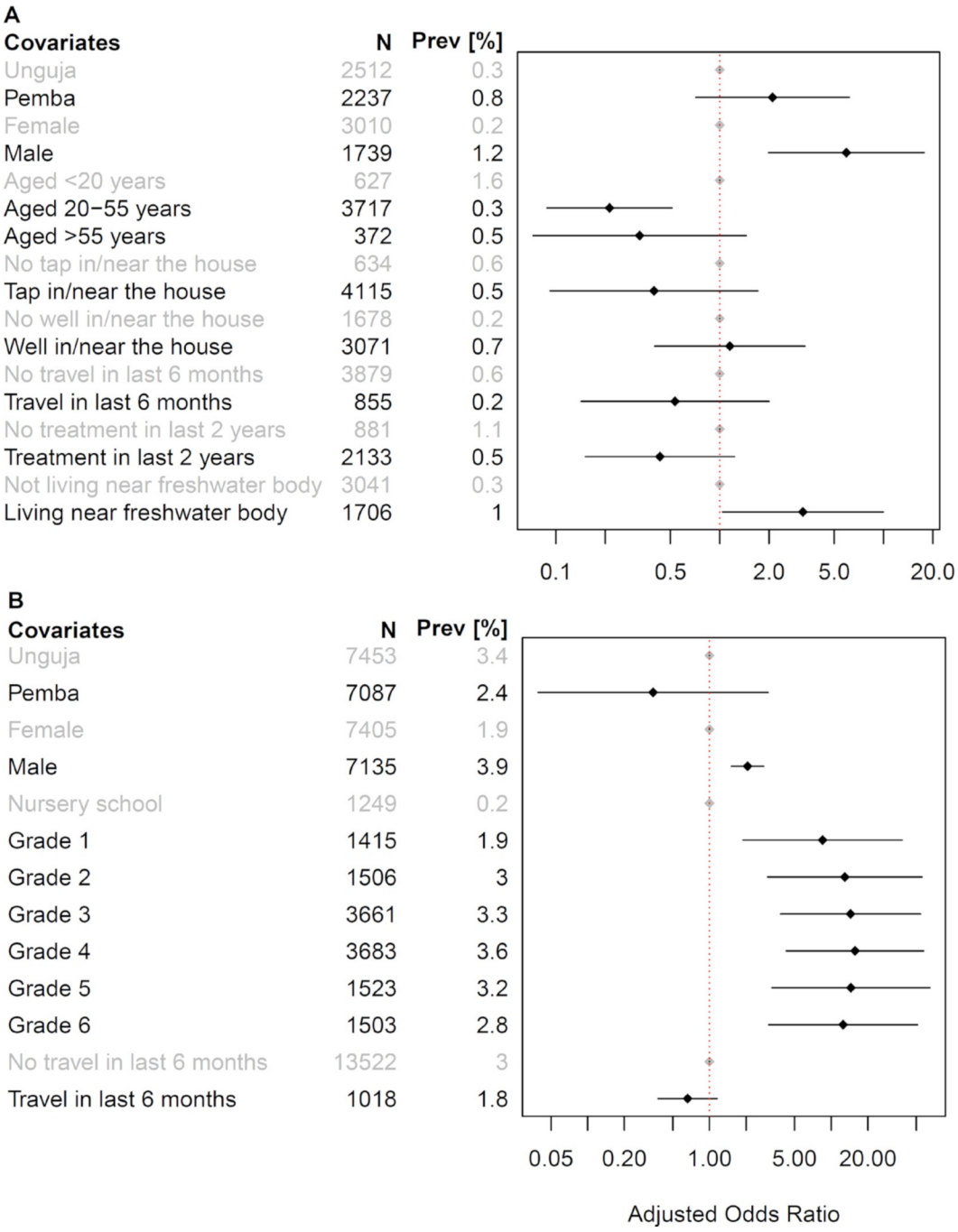


Fig 6. Multivariable analysis of risk factors for *S. haematobium* infections in adults (A) and schoolchildren (B) in Zanzibar in 2020. The figure shows the odds ratios for a *S. haematobium* infection adjusted for different risk factors. Grey dots indicate the reference categories. N: total number of participants analyzed per group; Prev [%]: *S. haematobium* prevalence per group.

https://doi.org/10.1371/journal.pntd.0009127.g006

of 4.0% were observed in schoolchildren in 2019. In 2020, the considerable rebound in the overall prevalence and also infection intensity was caused by certain hotspot areas, while most areas had very low prevalences throughout the study period and also after the 16-month treatment gap. The hotspot areas showed an unstable and undulating *S. haematobium* prevalence

pattern over the years from 2011/12 to 2020. Rapid reinfection after praziquantel treatment and the persistence of hotspots are well-known challenges for the sustainable control and elimination of schistosomiasis [4,30–33].

Most of the hotspot areas in Pemba and Unguja were not geographically isolated but neighbored with other hotspot areas. Clustering of *S. haematobium* infections in specific areas has also been observed in other studies, for example in Kenya [30,34]. The likelihood of reinfection is strongly dependent on the environment of regions and the force of transmission [9,35,36]. For example, an earlier study in some of the hotspot areas in Unguja conducted in 2014 showed that there were considerably more natural freshwater bodies in total and also more natural fresh water bodies containing the intermediate host snail transmitting *S. haematobium* compared with investigated low-prevalence areas [37].

In our study, the rebound in *S. haematobium* (re)infections from 2019 to 2020 was most prominent in children aged 8–16 year-old, and in boys. The latter observation is in line with another study from Senegal, which reports a higher *S. haematobium* reinfection rate in boys compared with girls after treatment with praziquantel [38]. However, the full age-profile of *Schistosoma* infections remains poorly researched, particularly in populations that received several rounds of MDA. We showed that in 2019, after 11 rounds of MDA in Zanzibar, infection levels across all age groups from four to 80 years were extremely low and a "real" age-prevalence distribution was inexistent. The "typical" age-prevalence curve, reported in reviews with a peak of *S. haematobium* infections occurring in children aged 8–15 years and a subsequent decrease and stabilization of a low prevalence in adult age [39,40], reformed quickly in the 16-month treatment gap in Zanzibar until early 2020.

Our risk factor analyses in 2020 indicated that men and boys had significantly higher odds of a *S. haematobium* infection than women and girls (OR: 6.24 and OR: 2.06, respectively). The observations that male individuals are more likely to harbor schistosomiasis is in line with previous surveys in Zanzibar and elsewhere [18,41–44]. Male individuals may be more engaged in behaviors that expose them to *S. haematobium* contaminated freshwater, such as swimming, playing and fishing [41,45,46]. In Pemba and Unguja, where Islam is the predominant religion, men also often use the rivers and ponds for ablution before their prayers. School aged children might get in frequent contact with contaminated freshwater when doing household chores such as washing dishes or clothes at the rivers and ponds near their home, or while engaging in leisure activities such as swimming and playing [47,48]. While boys might expose themselves more frequently to water than girls and thus acquire infections more often and quickly, we also observed that in line with their religious beliefs, girls frequently keep their clothes on while bathing or swimming, which might impose a certain barrier for cercarial penetration.

The risk factor analysis of adults in the CBS showed significantly elevated odds of *S. haematobium* infection for individuals who reported to live in close proximity to natural freshwater bodies (OR: 2.90). This observation confirms earlier reports from Zanzibar, where the proximity of households or schools to freshwater bodies containing the intermediate host snails was associated with *S. haematobium* infections [37,45]. Travel outside the home island, for example to the sister island, or mainland Tanzania, or other African countries, was not a significant risk factor in our study. Hence, at least to date, importation of urogenital schistosomiasis to Zanzibar does not seem to be a challenge and threat for elimination efforts. This finding is in contrast to reports from the neighboring island Mafia, where urogenital schistosomiasis is considered as an imported disease [49]. Also, access to clean water by having a tap or well near or at the home was not significantly associated with a *S. haematobium* infection in our study. This is in contrast to assumptions that access to clean water can improve the situation in endemic countries and reduce exposure to schistosomes [42,50]. However, one has to

keep in mind that many drivers for infection are complex, interconnected and multidirectional and that a systems epidemiology approach is necessary to put them into context [51].

A limitation of our large-scale community- and school-based study that involved more than 10,000 people annually is the yet small number of individuals and particularly adults that were diagnosed *S. haematobium*-positive. While this is an expected impediment when working in an elimination setting, the low infection numbers rendered the risk factor assessment difficult and resulted in huge relative differences in the *S. haematobium* prevalence between some years. Moreover, we used basic parasitological methods (reagent strips and urine filtration microscopy) to detect *S. haematobium* infections, which are not very sensitively detecting very light intensity infections [52]. Hence, many more people than identified might have carried a very light intensity infection, thereby biasing associations and resulting in an underestimation of the true prevalence.

Zanzibar has made great achievements towards schistosomiasis elimination over the past years applying population-based interventions. The current evidence that the *S. haematobium* prevalence and morbidity rebound quickly over the period of a 16-month treatment gap in hotspot areas but remain low in areas with a weaker force of transmission, emphasizes the need to rethink carefully future elimination strategy approaches. Clearly, due to the spatial heterogeneity of *S. haematobium* infections in Zanzibar and the varying risk of reinfection, future interventions aiming for interruption of *S. haematobium* transmission need to consider the micro-epidemiology of the islands and be adapted to it [19]. We hence suggest that interventions targeting hotspot areas in Zanzibar should be re-intensified and ideally include biannual MDA, snail control, behavior change and, in a multi-sectoral approach, improved access to safe water and sanitation. In low prevalence areas, future efforts might shift from MDA towards surveillance-response, including risk-based test-and-treat approaches using new and sensitive point-of-care diagnostic tools, xenomonitoring, focal snail control and health communication to ensure the identification and treatment of those infected but to avoid overtreatment and treatment fatigue of the healthy population. Setting up an effective surveillance-response system in areas that are or become low prevalence areas, will also help to sustain the gains made by reacting to any outbreak and sign of recrudescence in time and thus to ultimately progress towards a sustained interruption of *S. haematobium* transmission across Zanzibar. Our results also indicate that interruption of MDA and potentially other interventions, for example caused by a lack of donor funding, procurement issues, or the current Covid-19 pandemic, can result in a rapid resurgence of transmission and disease that needs to be reacted to in time. Finally, our study highlights the need for more research on the sustainability of the gains of control and on the criteria and conditions required for stopping schistosomiasis MDA.

Supporting information

S1 Dataset. Data that support the findings of this study from 2018–2020.
(CSV)

S1 Dictionary. Data dictionary that explains the data collected from 2018–2020.
(TXT)

Acknowledgments

We are grateful to all the schoolchildren and adults who participated in the surveys over the years 2011/12 to 2020. We thank the dedicated field- and laboratory teams in Pemba and Unguja who contributed to this work.

Author Contributions

Conceptualization: Jan Hattendorf, Said Mohammed Ali, Fatma Kabole, David Rollinson, Stefanie Knopp.

Data curation: Lydia Trippler, Stefanie Knopp.

Formal analysis: Lydia Trippler, Jan Hattendorf, Stefanie Knopp.

Funding acquisition: David Rollinson, Stefanie Knopp.

Investigation: Lydia Trippler, Shaali Makame Ame, Saleh Juma, Salum Abubakar, Said Mohammed Ali, Fatma Kabole, David Rollinson, Stefanie Knopp.

Methodology: Lydia Trippler, Shaali Makame Ame, Saleh Juma, Salum Abubakar, Said Mohammed Ali, Fatma Kabole, Stefanie Knopp.

Project administration: Shaali Makame Ame, Said Mohammed Ali, Fatma Kabole, David Rollinson, Stefanie Knopp.

Resources: Said Mohammed Ali, Fatma Kabole, David Rollinson.

Supervision: Shaali Makame Ame, Said Mohammed Ali, Fatma Kabole, David Rollinson, Stefanie Knopp.

Visualization: Lydia Trippler, Jan Hattendorf, Stefanie Knopp.

Writing – original draft: Lydia Trippler, Stefanie Knopp.

Writing – review & editing: Lydia Trippler, Shaali Makame Ame, Jan Hattendorf, Saleh Juma, Salum Abubakar, Said Mohammed Ali, Fatma Kabole, David Rollinson, Stefanie Knopp.

References

1. GBD 2017 DALYs and HALE Collaborators. Global, regional, and national disability-adjusted life-years (DALYs) for 359 diseases and injuries and healthy life expectancy (HALE) for 195 countries and territories, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet. 2018; 392 (10159):1859–922. Epub 2018/11/13. https://doi.org/10.1016/S0140-6736(18)32335-3 PMID: 30415748
2. WHO. Schistosomiasis key facts 2020 [cited 2020 12.04.2020]. Available from: https://www.who.int/news-room/fact-sheets/detail/schistosomiasis.
3. WHO. Ending the neglect to attain the Sustainable Development Goals—A road map for neglected tropical diseases 2021–2030. World Health Organization, 2020.
4. Deol AK, Fleming FM, Calvo-Urbano B, Walker M, Bucumi V, Gnandou I, et al. Schistosomiasis—assessing progress toward the 2020 and 2025 global goals. N Engl J Med. 2019; 381(26):2519–28. https://doi.org/10.1056/NEJMoa1812165 PMID: 31881138
5. Toor J, Rollinson D, Turner HC, Gouvras A, King CH, Medley GF, et al. Achieving elimination as a public health problem for *Schistosoma mansoni* and *S. haematobium*: When is community-wide treatment required? J Infect Dis. 2020; 221(Supplement_5):S525–s30. Epub 2019/12/13. https://doi.org/10.1093/infdis/jiz609 PMID: 31829414
6. Knopp S, Person B, Ame SM, Ali SM, Hattendorf J, Juma S, et al. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: a cluster-randomised trial. Lancet Glob Health. 2019; 7(8):e1118–e29. Epub 2019/07/01. https://doi.org/10.1016/S2214-109X(19)30189-5 PMID: 31255591
7. Allan F, Ame SM, Tian-Bi YT, Hofkin BV, Webster BL, Diakité NR, et al. Snail-related contributions from the Schistosomiasis Consortium for Operational Research and Evaluation program including xenomonitoring, focal mollusciciding, biological control, and modeling. Am J Trop Med Hyg. 2020; 103 (1_Suppl):66–79. Epub 2020/05/14. https://doi.org/10.4269/ajtmh.19-0831 PMID: 32400353
8. Pennance T, Archer J, Lugli EB, Rostron P, Llanwarne F, Ali SM, et al. Development of a molecular snail xenomonitoring assay to detect *Schistosoma haematobium* and *Schistosoma bovis* infections in their *Bulinus* snail hosts. Molecules. 2020; 25(17). Epub 2020/09/06. https://doi.org/10.3390/molecules25174011 PMID: 32887445

9. Gurarie D, Lo NC, Ndeffo-Mbah ML, Durham DP, King CH. The human-snail transmission environment shapes long term schistosomiasis control outcomes: Implications for improving the accuracy of predictive modeling. PLoS Negl Trop Dis. 2018; 12(5):e0006514. Epub 2018/05/22. https://doi.org/10.1371/journal.pntd.0006514 PMID: 29782500
10. Mgeni AF, Kisumku UM, McCullough FS, Dixon H, Yoon SS, Mott KE. Metrifonate in the control of urinary schistosomiasis in Zanzibar. Bull World Health Organ. 1990; 68(6):721–30. Epub 1990/01/01. PMID: 2127381
11. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba Island: programme organization and management. Trop Med Parasitol. 1989; 40(2):189–94. Epub 1989/06/01. PMID: 2505381
12. Albonico M, Chwaya HM, Montresor A, Stolfzfus RJ, Tielsch JM, Alawi KS, et al. Parasitic infections in Pemba Island school children. East Afr Med J. 1997; 74(5):294–8. Epub 1997/05/01. PMID: 9337006
13. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba Island; selective population chemotherapy of schoolchildren with haematuria to identify high-risk localities. Trans R Soc Trop Med Hyg. 1989; 83(6):805–10. Epub 1989/11/01. https://doi.org/10.1016/0035-9203(89)90336-2 PMID: 2515637
14. Stothard JR, Mook P, Mgeni AF, Khamis IS, Khamis AN, Rollinson D. Control of urinary schistosomiasis on Zanzibar (Unguja Island): a pilot evaluation of the educational impact of the Juma na Kichocho health booklet within primary schools. Mem Inst Oswaldo Cruz. 2006; 101 Suppl 1:119–24. Epub 2007/02/20.
15. Stothard JR, French MD, Khamis IS, Basanez MG, Rollinson D. The epidemiology and control of urinary schistosomiasis and soil-transmitted helminthiasis in schoolchildren on Unguja Island, Zanzibar. Trans R Soc Trop Med Hyg. 2009; 103(10):1031–44. Epub 2009/05/05. https://doi.org/10.1016/j.trstmh.2009.03.024 PMID: 19409588
16. Guidi A, Andolina C, Makame Ame S, Albonico M, Cioli D, Juma Haji H. Praziquantel efficacy and long-term appraisal of schistosomiasis control in Pemba Island. Trop Med Int Health. 2010; 15(5):614–8. Epub 2010/03/11. https://doi.org/10.1111/j.1365-3156.2010.02488.x PMID: 20214757
17. Knopp S, Mohammed KA, Ali SM, Khamis IS, Ame SM, Albonico M, et al. Study and implementation of urogenital schistosomiasis elimination in Zanzibar (Unguja and Pemba islands) using an integrated multidisciplinary approach. BMC Public Health. 2012; 12:930. Epub 2012/11/01. https://doi.org/10.1186/1471-2458-12-930 PMID: 23110494
18. Knopp S, Person B, Ame SM, Mohammed KA, Ali SM, Khamis IS, et al. Elimination of schistosomiasis transmission in Zanzibar: baseline findings before the onset of a randomized intervention trial. PLoS Negl Trop Dis. 2013; 7(10):e2474. Epub 2013/10/23. https://doi.org/10.1371/journal.pntd.0002474 PMID: 24147165
19. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-Year intervention study on elimination of urogenital schistosomiasis in Zanzibar: Parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019; 13(5):e0007268. Epub 2019/05/07. https://doi.org/10.1371/journal.pntd.0007268 PMID: 31059495
20. Office of Chief Government Statistician Zanzibar. Zanzibar statistical abstract 2019. 2020.
21. Office of Chief Government Statistician Zanzibar. Zanzibar in figures 2019. 2020.
22. Office of Chief Government Statistician Zanzibar. Household budget survey 2014/2015. 2016.
23. Ministry of Health Tanzania. Tanzania demographic and health survey and malaria indicator survey 2015–2016—Final report. 2016.
24. WHO. Prevention and control of schistosomiasis and soil-transmitted helminthiasis: report of a WHO expert committee. World Health Organization, 2002.
25. Montresor A, Engels D, Ramsan M, Foum A, Savioli L. Field test of the 'dose pole' for praziquantel in Zanzibar. Trans R Soc Trop Med Hyg. 2002; 96(3):323–4. Epub 2002/08/15. https://doi.org/10.1016/s0035-9203(02)90111-2 PMID: 12174788
26. Montresor A, Crompton D, Hall A, Bundy D, Savioli L. Guidelines for the evaluation of soil-transmitted helminthiasis and schistosomiasis at community level. World Health Organization, 1998.
27. Krauth SJ, Greter H, Stete K, Coulibaly JT, Traoré SI, Ngandolo BN, et al. All that is blood is not schistosomiasis: experiences with reagent strip testing for urogenital schistosomiasis with special consideration to very-low prevalence settings. Parasit Vectors. 2015; 8:584. Epub 2015/11/12. https://doi.org/10.1186/s13071-015-1165-y PMID: 26554822
28. Tomson C, Porter T. Asymptomatic microscopic or dipstick haematuria in adults: which investigations for which patients? A review of the evidence. BJU Int. 2002; 90(3):185–98. Epub 2002/07/23. https://doi.org/10.1046/j.1464-410x.2002.02841.x PMID: 12133052
29. McDonald MM, Swagerty D, Wetzel L. Assessment of microscopic hematuria in adults. Am Fam Physician. 2006; 73(10):1748–54. Epub 2006/06/01. PMID: 16734050

30. Kittur N, King CH, Campbell CH, Kinung'hi S, Mwinzi PNM, Karanja DMS, et al. Persistent hotspots in Schistosomiasis Consortium for Operational Research and Evaluation studies for gaining and sustaining control of schistosomiasis after four years of mass drug administration of praziquantel. Am J Trop Med Hyg. 2019; 101(3):617–27. Epub 2019/07/10. https://doi.org/10.4269/ajtmh.19-0193 PMID: 31287046

31. Wiegand RE, Mwinzi PNM, Montgomery SP, Chan YL, Andiego K, Omedo M, et al. A persistent hotspot of *Schistosoma mansoni* infection in a five-year randomized trial of praziquantel preventative chemotherapy strategies. J Infect Dis. 2017; 216(11):1425–33. Epub 2017/10/03. https://doi.org/10.1093/infdis/jix496 PMID: 28968877

32. Elmorshedy H, Bergquist R, El-Ela NE, Eassa SM, Elsakka EE, Barakat R. Can human schistosomiasis mansoni control be sustained in high-risk transmission foci in Egypt? Parasit Vectors. 2015; 8:372. Epub 2015/07/16. https://doi.org/10.1186/s13071-015-0983-2 PMID: 26174621

33. Dejon-Agobé JC, Edoa JR, Honkpehedji YJ, Zinsou JF, Adégbitè BR, Ngwese MM, et al. *Schistosoma haematobium* infection morbidity, praziquantel effectiveness and reinfection rate among children and young adults in Gabon. Parasit Vectors. 2019; 12(1):577. https://doi.org/10.1186/s13071-019-3836-6 PMID: 31823806

34. Chadeka EA, Nagi S, Sunahara T, Cheruiyot NB, Bahati F, Ozeki Y, et al. Spatial distribution and risk factors of *Schistosoma haematobium* and hookworm infections among schoolchildren in Kwale, Kenya. PLoS Negl Trop Dis. 2017; 11(9):e0005872–e. https://doi.org/10.1371/journal.pntd.0005872 PMID: 28863133

35. Kittur N, Binder S, Campbell CH, King CH, Kinung'hi S, Olsen A, et al. Defining persistent hotspots: areas that fail to decrease meaningfully in prevalence after multiple years of mass drug administration with praziquantel for control of schistosomiasis. Am J Trop Med Hyg. 2017; 97(6):1810–7. Epub 2017/10/11. https://doi.org/10.4269/ajtmh.17-0368 PMID: 29016344

36. Mari L, Ciddio M, Casagrandi R, Perez-Saez J, Bertuzzo E, Rinaldo A, et al. Heterogeneity in schistosomiasis transmission dynamics. J Theor Biol. 2017; 432:87–99. https://doi.org/10.1016/j.jtbi.2017.08.015 PMID: 28823529

37. Pennance T, Person B, Muhsin MA, Khamis AN, Muhsin J, Khamis IS, et al. Urogenital schistosomiasis transmission on Unguja Island, Zanzibar: Characterisation of persistent hot-spots. Parasit Vectors. 2016; 9(1):646. Epub 2016/12/18. https://doi.org/10.1186/s13071-016-1847-0 PMID: 27986092

38. Senghor B, Diaw OT, Doucoure S, Sylla SN, Seye M, Talla I, et al. Efficacy of praziquantel against urinary schistosomiasis and reinfection in Senegalese school children where there is a single well-defined transmission period. Parasit Vectors. 2015; 8(1):362. https://doi.org/10.1186/s13071-015-0980-5 PMID: 26156522

39. Gryseels B, Polman K, Clerinx J, Kestens L. Human schistosomiasis. Lancet. 2006; 368(9541):1106–18. Epub 2006/09/26. https://doi.org/10.1016/S0140-6736(06)69440-3 PMID: 16997665

40. Colley DG, Secor WE. Immunology of human schistosomiasis. Parasite Immunol. 2014; 36(8):347–57. Epub 2014/08/22. https://doi.org/10.1111/pim.12087 PMID: 25142505

41. Lee Y-H, Lee J-S, Jeoung H-G, Kwon I-S, Mohamed AAWS, Hong S-T. Epidemiological survey on schistosomiasis and intestinal helminthiasis among village residents of the rural river basin area in White Nile State, Sudan. Korean J Parasitol. 2019; 57(2):135–44. Epub 2019/04/30. https://doi.org/10.3347/kjp.2019.57.2.135 PMID: 31104405

42. Kapito-Tembo AP, Mwapasa V, Meshnick SR, Samanyika Y, Banda D, Bowie C, et al. Prevalence distribution and risk factors for *Schistosoma hematobium* infection among school children in Blantyre, Malawi. PLoS Negl Trop Dis. 2009; 3(1):e361. https://doi.org/10.1371/journal.pntd.0000361 PMID: 19156193

43. Nagi S, Chadeka EA, Sunahara T, Mutungi F, Justin YK, Kaneko S, et al. Risk factors and spatial distribution of *Schistosoma mansoni* infection among primary school children in Mbita District, Western Kenya. PLoS Negl Trop Dis. 2014; 8(7):e2991. Epub 2014/07/25. https://doi.org/10.1371/journal.pntd.0002991 PMID: 25058653

44. Atalabi TE, Lawal U, Ipinlaye SJ. Prevalence and intensity of genito-urinary schistosomiasis and associated risk factors among junior high school students in two local government areas around Zobe Dam in Katsina State, Nigeria. Parasit Vectors. 2016; 9(1):388. https://doi.org/10.1186/s13071-016-1672-5 PMID: 27388007

45. Rudge JW, Stothard JR, Basanez MG, Mgeni AF, Khamis IS, Khamis AN, et al. Micro-epidemiology of urinary schistosomiasis in Zanzibar: Local risk factors associated with distribution of infections among schoolchildren and relevance for control. Acta Trop. 2008; 105(1):45–54. Epub 2007/11/13. https://doi.org/10.1016/j.actatropica.2007.09.006 PMID: 17996207

46. Geleta S, Alemu A, Getie S, Mekonnen Z, Erko B. Prevalence of urinary schistosomiasis and associated risk factors among Abobo Primary School children in Gambella Regional State, southwestern

Ethiopia: a cross sectional study. Parasit Vectors. 2015; 8(1):215. https://doi.org/10.1186/s13071-015-0822-5 PMID: 25886292

47. Angora EK, Boissier J, Menan H, Rey O, Tuo K, Touré AO, et al. Prevalence and risk factors for schistosomiasis among schoolchildren in two settings of Côte d'Ivoire. Trop Med Infect Dis. 2019; 4(3):110. https://doi.org/10.3390/tropicalmed4030110 PMID: 31340504

48. Person B, Ali SM, A'Kadir FM, Ali JN, Mohammed UA, Mohammed KA, et al. Community knowledge, perceptions, and practices associated with urogenital schistosomiasis among school-aged children in Zanzibar, United Republic of Tanzania. PLoS Negl Trop Dis. 2016; 10(7):e0004814. Epub 2016/07/12. https://doi.org/10.1371/journal.pntd.0004814 PMID: 27399310

49. Stothard JR, Ameri H, Khamis IS, Blair L, Nyandindi US, Kane RA, et al. Parasitological and malacological surveys reveal urogenital schistosomiasis on Mafia Island, Tanzania to be an imported infection. Acta Trop. 2013; 128(2):326–33. Epub 2012/09/27. https://doi.org/10.1016/j.actatropica.2012.09.006 PMID: 23010161

50. Grimes JET, Croll D, Harrison WE, Utzinger J, Freeman MC, Templeton MR. The relationship between water, sanitation and schistosomiasis: A systematic review and meta-analysis. PLoS Negl Trop Dis. 2014; 8(12):e3296. https://doi.org/10.1371/journal.pntd.0003296 PMID: 25474705

51. Krauth SJ, Balen J, Gobert GN, Lamberton PHL. A call for systems epidemiology to tackle the complexity of schistosomiasis, its control, and its elimination. Trop Med Infect Dis. 2019; 4(1):21.

52. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Urogenital schistosomiasis elimination in Zanzibar: accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections. Parasit Vectors. 2018; 11(1):552. Epub 2018/10/26. https://doi.org/10.1186/s13071-018-3136-6 PMID: 30352631

3.3. Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study

Lydia Trippler[1,2], Jan Hattendorf[1,2], Said Mohammed Ali[3], Shaali Makame Ame[3], Saleh Juma[4], Fatma Kabole[4], Stefanie Knopp[1,2]

1 Swiss Tropical and Public Health Institute, Basel, Switzerland

2 University of Basel, Basel, Switzerland

3 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

4 Neglected Diseases Programme, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

This article was published in:

BMC Infectious Diseases (2021), Sep 30; 21(1):1024

doi: 10.1186/s12879-021-06620-8

BMC Infectious Diseases

STUDY PROTOCOL Open Access

Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study

Lydia Trippler[1,2], Jan Hattendorf[1,2], Said Mohammed Ali[3], Shaali Makame Ame[3], Saleh Juma[4], Fatma Kabole[4] and Stefanie Knopp[1,2*]

Abstract

Background: Global elimination of schistosomiasis as a public health problem is set as target in the new World Health Organization's Neglected Tropical Diseases Roadmap for 2030. Due to a long history of interventions, the Zanzibar islands of Tanzania have reached this goal since 2017. However, challenges occur on the last mile towards interruption of transmission. Our study will investigate new tools and strategies for breaking schistosomiasis transmission.

Methods: The study is designed as an intervention study, documented through repeated cross-sectional surveys (2020–2024). The primary endpoint will be the sensitivity of a surveillance-response approach to detect and react to outbreaks of urogenital schistosomiasis over three years of implementation. The surveys and multi-disciplinary interventions will be implemented in 20 communities in the north of Pemba island. In low-prevalence areas, surveillance-response will consist of active, passive and reactive case detection, treatment of positive individuals, and focal snail control. In hotspot areas, mass drug administration, snail control and behaviour change interventions will be implemented. Parasitological cross-sectional surveys in 20 communities and their main primary schools will serve to adapt the intervention approach annually and to monitor the performance of the surveillance-response approach and impact of interventions. *Schistosoma haematobium* infections will be diagnosed using reagent strips and urine filtration microscopy, and by exploring novel point-of-care diagnostic tests.

Discussion: Our study will shed light on the field applicability and performance of novel adaptive intervention strategies, and standard and new diagnostic tools for schistosomiasis elimination. The evidence and experiences generated by micro-mapping of *S. haematobium* infections at community level, micro-targeting of new adaptive intervention approaches, and application of novel diagnostic tools can guide future strategic plans for schistosomiasis elimination in Zanzibar and inform other countries aiming for interruption of transmission.

Trial registration ISRCTN, ISCRCTN91431493. Registered 11 February 2020, https://www.isrctn.com/ISRCTN91431493

Keywords: Adaptive interventions, Behaviour change, *Bulinus*, Case finding, Diagnostics, Elimination, Hotspot, Interruption of transmission, Schistosomiasis, *Schistosoma haematobium*, Snail control, Surveillance-response, Zanzibar

Background

Schistosomiasis is a neglected tropical disease (NTD) with a considerable impact on global health [1, 2]. Since the mid-1980s, efforts in endemic countries mainly focused on the control of morbidity using preventive

*Correspondence: s.knopp@swisstph.ch
[1] Swiss Tropical and Public Health Institute, Socinstrasse 57, 4051 Basel, Switzerland
Full list of author information is available at the end of the article

chemotherapy with praziquantel [3]. A paradigm shift occurred in recent years: in 2012, the World Health Organization (WHO) declared as goal to eliminate schistosomiasis as a public health problem and to interrupt transmission in selected areas by 2025 [4]. In the new WHO Roadmap on NTDs published in 2021, the global elimination of schistosomiasis as a public health problem and the validated absence of infections in humans in 25 among 78 endemic countries are set as targets for 2030 [5].

There are several countries and areas that have made great progress in the fight against schistosomiasis in the past and may aim to achieve full interruption of transmission in the next few years [6–10]. These countries will need clear guidance on which intervention strategies to apply, which population groups to target, which diagnostics to use, and at what thresholds to change and adapt their strategies [11–13]. Moving from morbidity control towards elimination as public health problem and interruption of transmission, WHO recommends in their schistosomiasis progress report 2001–2011 and strategic plan 2012–2020, the intensification of mass drug administration (MDA) and the implementation of complementary public-health interventions in addition to preventive chemotherapy (Fig. 1) [4]. In World Health Assembly resolution 65.21 (05/12) these complementary interventions are indicated as "strengthened health systems, […] provision of water and sanitation, as well as hygiene education and snail control" [14]. In the new WHO NTD Roadmap 2021–2030, besides MDA, the following core strategic interventions against schistosomiasis are listed: Water, Sanitation and Hygiene (WASH), vector control, veterinary public health, case management and other interventions such as behaviour change, self-care and environmental management [5]. Finally, according to WHO, once interruption of transmission is close or has been achieved, affected countries will need to implement a surveillance system "to detect and respond to resurgence of transmission and to prevent reintroduction from regions where the disease is still endemic" [4]. Despite these recommendations, specific guidance and thresholds on when, where and how to adapt intervention strategies in near-to-elimination settings is yet to be developed [11, 12, 15]. More evidence on the feasibility, impact, effectiveness and sustainability of multi-pronged intervention approaches needs to be generated.

A wealth of experience and insights regarding multidisciplinary interventions and research for urogenital schistosomiasis elimination was gained over the past decade from 2011 to 2020 within the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) project [6, 16–19]. The islands Pemba and Unguja, belonging to the Zanzibar archipelago of the United Republic of Tanzania, used to be highly endemic for urogenital schistosomiasis in the past century [20–22]. To combat urogenital schistosomiasis, caused by *Schistosoma haematobium*, on the islands, regular MDA campaigns with praziquantel administration to schoolchildren and the whole community were implemented since the 1980s [23–26]. In 2010, Zanzibar had managed to reach low prevalences of *S. haematobium* infections and subsequently committed to eliminate urogenital schistosomiasis from the islands [17, 18]. From 2012 till 2017, in addition to biannual MDA, some randomised areas received interventions with chemical molluscicide to target *Bulinus globosus,* the intermediate host snail for *S. haematobium* in Zanzibar [27, 28] and behaviour change measures to reduce people`s freshwater contact [17, 19]. In 2017, most areas on the Zanzibar islands had eliminated urogenital schistosomiasis as a public health problem (defined as < 1% heavy intensity infections) [5, 17, 18]. However, transmission of *S. haematobium* infections was not yet interrupted and several challenges were identified on the way towards schistosomiasis elimination.

These challenges included a marked temporal and spatial heterogeneity of *S. haematobium* prevalence across the islands, as well as the application of standard diagnostic tests with insufficient sensitivity to detect light intensity infections [6, 17, 18, 29–32]. While most of the study sites showed continuously low prevalences from 2011/12 to 2020, some "hotspot" areas with consistent or recurring moderate or high prevalences existed [6, 17, 18]. To address this heterogeneity and to sustain and accelerate the gains made towards elimination, there is

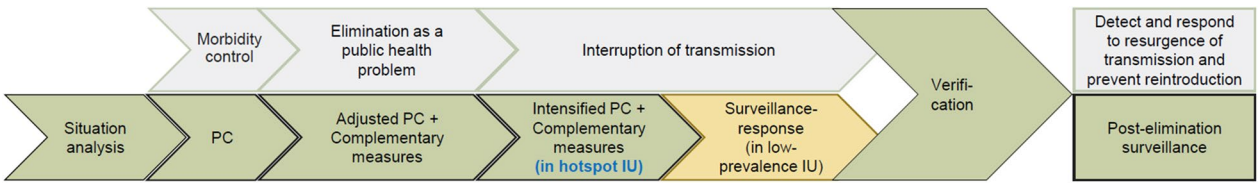


Fig. 1 Programmatic steps to control and eliminate schistosomiasis. In green and gray text boxes: recommendations according to WHO [4]; in blue fonts and yellow text box: additional components of the SchistoBreak study. PC: preventive chemotherapy; IU: implementation unit

a need to target and regularly adapt interventions to the local micro-epidemiology [12, 17, 18].

In low-prevalence areas, it will be important to progress towards complete interruption of transmission by applying an intervention approach that enables the reliable identification and treatment of all cases and thus prevents outbreaks and recrudescence, without overtreating a mostly healthy population [18] and thereby risking treatment fatigue and/or the development of resistance against praziquantel [33]. The participation of countries in the development and use of surveillance-response approaches to ensure that the progress made is sustained and advanced has been stressed in several publications [15, 34–39].

In hotspot areas that are often characterized by a large number of water bodies containing the intermediate host snails and proximity of households and schools to transmission sites [40, 41] there is a need to tackle the persistent transmission to progress towards elimination using a comprehensive package of interventions, including MDA, but also other interventions such as snail control and behaviour change communication and WASH measures [11, 17, 18, 32].

Accurate, reliable and affordable diagnostic tools are an essential requirement for NTD programmes and have been identified as a priority area for critical action in the WHO NTD Roadmap 2021–2030 [5, 13, 15, 42, 43]. Moving towards schistosomiasis elimination, there is an enhanced need for sensitive and specific diagnostic tests that are high-throughput, and affordable and applicable at the point of care to assess reliably *S. haematobium* infections, prevalences and incidence [13, 43]. An accurate picture of the endemic situation is important for programmatic decision making, to tailor specific intervention packages in line with (yet to be developed) target thresholds to those in need, to determine correctly the performance and impact of interventions in elimination settings, and to document sustained elimination [13, 29–31, 44].

In our protocol, we describe an implementation research study that will address the focality and heterogeneity of *S. haematobium* transmission, using novel adaptive intervention strategies as well as standard and new diagnostic tools for schistosomiasis elimination in Zanzibar.

Methods/design

Study aim

The overall objective of the study is to investigate new tools and strategies for breaking schistosomiasis transmission.

Primary and secondary objectives

The primary objective of this study is to quantify the sensitivity of an adaptive surveillance-response approach for its ability to detect *S. haematobium* infected individuals in low-prevalence areas to trigger an appropriate intervention response. The primary outcome variable will be the number of *S. haematobium* infected individuals detected and reported through the surveillance approach divided by the number of positive individuals in the population as extrapolated from the cross-sectional surveys, i.e. the mean sensitivity of the surveillance-response approach determined over 3 years.

Secondary outcome analyses will cover additional performance parameters of the surveillance approach, as defined by WHO [45]. Moreover, we will assess the impact of multi-disciplinary interventions in hotspot areas, as well as the coverage of test-and-treat activities in low-prevalence areas and of MDA in hotspot areas. Other secondary outcomes will include the accuracy of several diagnostic approaches and micro-mapping of characteristics of the study shehias, including the number and location of schools, madrassas (Islamic schools), and health facilities, the number and location of water bodies and the abundance of intermediate host snails of the genus *Bulinus*.

Study design

The study is designed as intervention study, documented through repeated cross-sectional surveys to assess the performance of the surveillance-response approach.

Study setting

The implementation research study with the acronym "SchistoBreak" is a joint project with partners from the Swiss Tropical and Public Health Institute (Swiss TPH), Basel, Switzerland the Public Health Laboratory-Ivo de Carneri (PHL-IdC), Pemba, United Republic of Tanzania and the Neglected Diseases Programme of the Ministry of Health, Social Welfare, Elderly, Gender, and Children (MoHSWEGC) Zanzibar, United Republic of Tanzania. The fieldwork for the SchistoBreak study will be conducted in Pemba, an island that forms part of the Zanzibar archipelago, from March 2020 to June 2024. The population density of Pemba was 530 people/km^2 in 2012 and the projected population for 2019 was around 500,000 people [46]. The study area will consist of 20 shehias (small administrative areas) located in the rural districts Micheweni and Wete in the north of Pemba (Fig. 2). The average population size in the study shehias is ~ 3,900 individuals per shehia [47]. To address the small-scale heterogeneity in the study area and to micro-target implementation, not the districts but each shehia

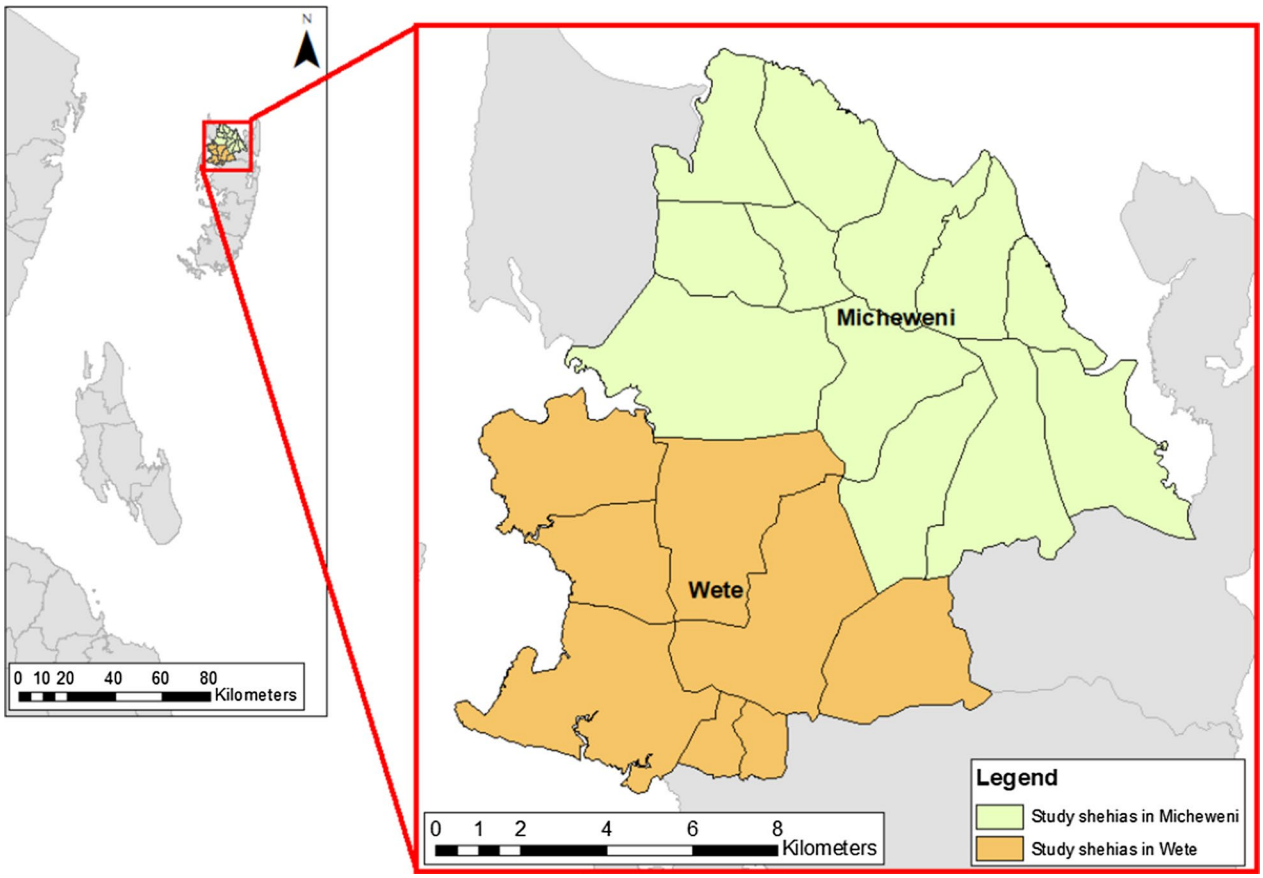


Fig. 2 Twenty study shehias (implementation unit) of the SchistoBreak project in Wete and Micheweni districts in Pemba, Tanzania. The image base map (United Republic of Tanzania – Subnational administrative boundaries) was downloaded from OCHA services (https://data.humdata.org/dataset/tanzania-administrative-boundaries-level-1-to-3-regions-districts-and-wards-with-2012-population). The data source is: Tanzania National Bureau of Statistics/UN OCHA ROSA. The data are published under the following license: Creative Commons Attribution for Intergovernmental Organisations (CC BY-IGO; (https://creativecommons.org/licenses/by/3.0/igo/legalcode)). Additionally, we received written permission to use and adapt the data from the UN Office for the Coordination of Humanitarian Affairs (OCHA). Additional shape files for the map (shehia boundaries) were provided by the Zanzibar Commission of Lands to the Zanzibar Neglected Diseases Programme

will serve as one implementation unit (IU). The 20 study shehias were selected based on their isolated but contiguous location in the north of Pemba and very good documentation of *S. haematobium* prevalences in 10 among the 20 shehias from 2012 to 2020 [6, 17, 18].

The study will take place in communities, schools and health facilities. In Zanzibar, public primary schools contain the grades 1–6 and some schools include a nursery school. In addition, most of the children in Zanzibar visit a madrassa [48]. The net enrolment rate in primary schools was 68.4% in Micheweni and 81.9% in Wete in 2014/15 [49].

Participant eligibility

All individuals who meet the following inclusion criteria are eligible to participate in the study:

1. All persons aged ≥ 4 years, living in the study shehias.
2. Submitted informed consent form (ICF) signed by parent or legal guardian in case of participating children and adolescents, or signed by the participant in case of participating adults, and submitted assent form in case of children aged 12 years and older signed by the participating children.
3. Urine sample with sufficient volume to perform diagnostic tests provided.

All individuals who do not meet the following exclusion criteria are not eligible to participate in the study:

1. Children < 4 years.
2. Children, adolescents and adults not living in the study area.

3. ICF not submitted or not signed by parent or legal guardian in case of participating children or adolescents or not signed by the participant in case of participating adults. Assent form not submitted or not signed by the child in case of participating children aged 12 years or older.
4. No urine sample of sufficient volume to perform diagnostic tests provided.

Shehia characteristics survey

Upon start of the SchistoBreak project, we will conduct a survey to gain an aggregate picture of the characteristics of the 20 study shehias. In a first step, in each shehia, we will meet with the sheha (head of shehia) and invite him to participate in a questionnaire interview to collect data about the population size, the number and type of schools, natural open freshwater bodies, health facilities and public clean water sources available in each shehia. Subsequently, with the sheha's permission and the support of an assistant sheha, we will visit all nursery, primary and secondary schools and all madrassas in the shehia and assess the geolocation, type of school, and the number of children enrolled in the school. Moreover, we will visit all known human water contact sites (HWCSs) at the natural open freshwater bodies in the shehias and collect data of locality, type and characteristics of the water body, regular behavioural activities at the water bodies and the abundance of freshwater snail species and specifically the intermediate host snail *Bulinus*. Finally, we will visit all health facilities in the shehias and determine the location and type of facility, i. e. whether they are of public or private status and offer mother and child health care services. For all processes of data collection during the shehia characteristics survey, we will use Open Data Kit (ODK) software (www.opendatakit.org), installed on a computer tablet (Samsung Galaxy Tab A 2019).

Annual cross-sectional parasitological surveys

The annual cross-sectional surveys will be implemented in all 20 communities and their main public primary schools at sub-district shehia level. This micro-mapping approach will allow us to determine *S. haematobium* prevalences in the school-aged population, which is at highest risk of schistosomiasis, as well as in the whole community to get an accurate picture of infection levels in preschool-aged children, school-aged children, adolescents and adults. The cross-sectional surveys will serve to stratify the study area into low-prevalence and hotspot IUs and hence to micro-target the interventions according to pre-set prevalence thresholds. Moreover, the cross-sectional surveys will allow to determine and monitor the performance of the surveillance-response approach and impact of interventions over the study period. Participants of the annual cross-sectional parasitological surveys identified as infected with *S. haematobium* will be offered treatment with praziquantel (40 mg/kg using a dose pole) [50].

Annual community-based parasitological surveys

Selection of houses and participants

For the annual community-based surveys, 70 housing structures per shehia will be selected by a computer-based randomisation procedure from Geographical Information System (GIS) shape files provided by the Zanzibar Mapping Initiative (http://www.zanzibarmapping.com) of the Commission of Lands Zanzibar. Randomly selected housing structures will be located by field enumerators by using the navigation app Maps.me (https://www.maps.me/) combined with ODK installed on a computer tablet (Samsung Galaxy Tab A 2019). Accounting for a 30% dropout of housing structures that may not be inhabited (i.e. shops, sheds, mosques, houses under construction or abandoned houses) and households without any member willing to participate, we estimate that we will have a final sample size of 50 houses per shehia included in the survey. People sharing the same kitchen or pot will define a household. All household members meeting the inclusion criteria will be invited to participate in the survey and to provide one own urine sample, which will be examined for *S. haematobium* infection markers. With an average of five people per household, we estimate to collect urine samples from a total of 250 participants per shehia. Additionally, one adult household member present at the first visit to the house will be invited to participate in a questionnaire survey to assess household characteristics. If more than one adult is present and eligible to participate at first visit, we will randomly select the participant of the questionnaire survey by using a playing-cards approach.

Data collection

A field enumerator using ODK and the wayfinding app Maps.me will visit each selected housing structure. The geolocation of each housing structure will be recorded in ODK. If the house is inhabited, the study will be explained in lay terms to the present household members and they will be invited to participate in the survey. Information and consent forms (ICF) for all eligible household members and assent forms for children aged 12 to 18 years will be distributed. Once adult participants and in the case of children aged below 18 years their parents provided written informed consent by

signing the form and, additionally, once children aged 12 to 18 years agreed to participate by signing the child assent form, plastic containers (100 ml) for urine collection will be distributed for each participant. The plastic containers will be labelled with a unique identifier code and an individual sticker picture, which both match the same identifier code and sticker picture on a provided paper form containing the household member names to support their correct identification. Additionally, one adult household member will be invited to answer several questions related to participation in the last MDA, water contact behaviour, access to safe water sources, opinion regarding intervention approaches against schistosomiasis, and the number of individuals living in the household and their main demographic information, such as sex and age. On the following day, the study team will revisit the households and collect the (remaining) signed ICFs, signed assent forms and filled urine containers. Each shehia will be visited for three days, to cover all selected houses and ensure maximal participation and compliance with urine collection.

School-based parasitological surveys

Selection of participants

The annual school-based cross sectional survey will be conducted in the main public primary school of each shehia. If a shehia has several public primary schools, the school with most students will be surveyed. If a shehia has no public primary school, no school will be surveyed. In each of the schools, a total of 175 students aged 4–17 years will be randomly selected for urine collection. For this purpose, one class of each nursery and standard 1–6 grades, will be selected based on computer-randomised lists. In the selected classes, all children will line up, stratified by sex. Subsequently, we will systematically select each third child in the lines to be included in the study until 25 children per class are reached. This procedure will be continued until we reach a total of 175 selected children from the 7 selected grades. Accounting for a 20% drop-out due to non-consenting parents, absenteeism of children or inability to produce a urine sample of sufficient volume, we aim for a final sample size of 20 children per standard (i.e. a total of 140 students per school).

Data collection

Each selected child will be provided with an ICF for their parents to read and sign. Each selected child aged 12 years or older will additionally be invited to sign an assent form on its own behalf. On the following day, once signed ICFs and assent forms are submitted, children will be registered, including information about their age, sex, travel history and participation in the last round of MDA. Subsequently, children will be handed over a plastic container labelled with a unique identifier code and invited to produce their own urine sample, which will be collected by the study team on the same morning. In addition, participants from grade 3, 4, and 5 will be invited to participate in a questionnaire interview about the knowledge, attitudes and practices regarding schistosomiasis transmission and prevention (KAP). The children will be asked about their knowledge of the animals that are part of the *S. haematobium* lifecycle, their own water contact behaviour, access to safe water sources, and their knowledge about opportunities to prevent both getting and spreading schistosomiasis.

Interventions and data collection

In the study, specific intervention packages will be tailored and adapted to the local micro-epidemiology of urogenital schistosomiasis in the shehias. Low-prevalence IUs will receive tailored surveillance-response measures, consisting of active, passive and reactive case finding, treatment of *S. haematobium*-positive individuals and focal snail control. Hotspot IUs will receive a multi-disciplinary intervention package including MDA, snail control and behaviour change communication. The classification of the study shehias into low-prevalence or hotspot IUs will be based on the results of annual cross-section surveys. Initially, a shehia will be considered a low-prevalence IU if it shows an apparent *S. haematobium* prevalence < 2% in the community-based survey and an apparent *S. haematobium* prevalence < 3% in the school-based survey, based on single urine filtration microscopy. Otherwise, the shehia will be considered a "hotspot" IU (Fig. 3). Results gained over the study period will show whether these thresholds and respective intervention packages successfully sustain and accelerate the gains and prevent recrudescence of *S. haematobium* transmission in low-prevalence IUs. The thresholds will be evaluated annually in light of the results of the cross-sectional surveys and adapted if needed.

Surveillance-response activities in low-prevalence implementation units

In a surveillance-response system, disease-related data are continuously and systematic collected and analysed. With constant monitoring, resurgence of transmission can be detected immediately and interventions can be implemented as a response to prevent reintroduction of the disease [4, 45].

In our low-prevalence IUs, where large parts of the population are not infected with *S. haematobium*, the intervention approach is shifted from large-scale

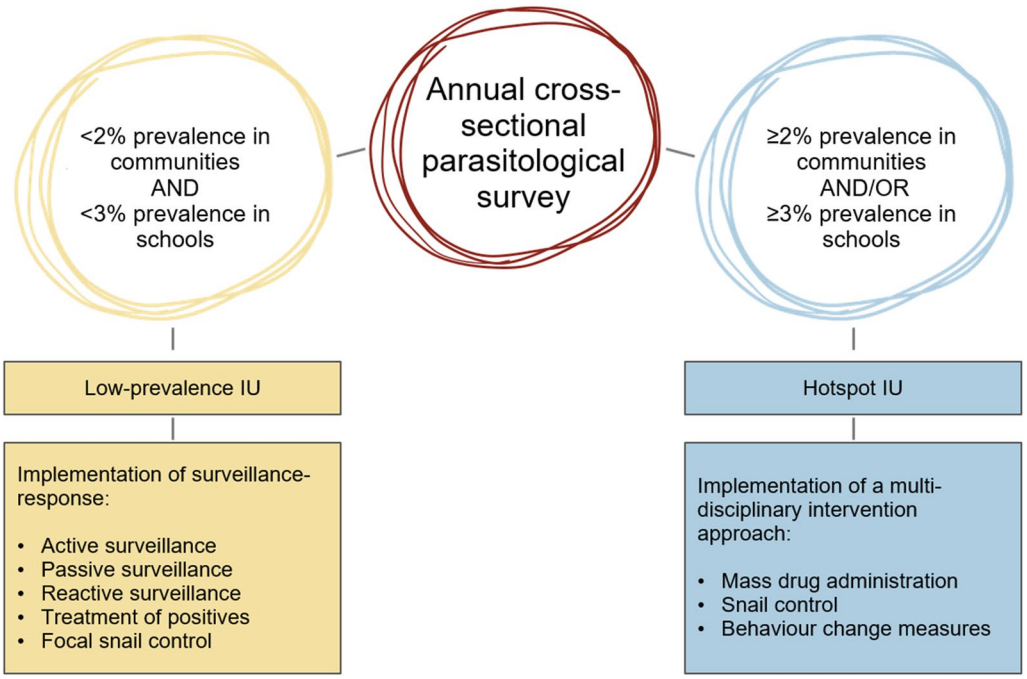


Fig. 3 Classification of the SchistoBreak study shehias in Pemba, Tanzania, into low-prevalence and hotspot implementation units (IUs) based on adaptive *S. haematobium* prevalence thresholds

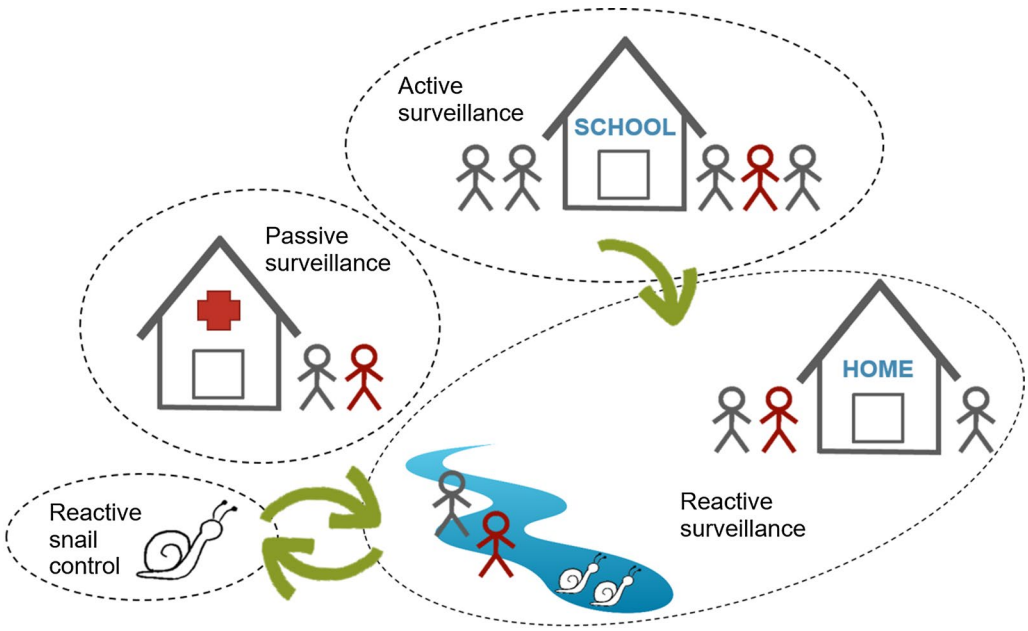


Fig. 4 Interventions and components of surveillance-response activities implemented in low-prevalence shehias of the SchistoBreak study in Pemba, Tanzania

measures with high coverage towards targeted micro-interventions, i.e. from MDA in communities and schools to risk-based surveillance-response, including active, passive and reactive case finding, treatment of haematuria-positive and/or *S. haematobium*-infected individuals, and reactive snail control (Fig. 4).

As starting point for risk-based surveillance activities, we will select the largest public primary school in each

shehia, and a madrassa with a minimum of 50 children that is located at maximum 500 m away from a water body and the farthest away from the primary school to gain a differentiated picture about the *S. haematobium* micro-epidemiology at school level in each shehia [12].

For active case finding, we will visit the selected schools at least once per year, register all children of the grades 3–5 present at the first day of the visit, and provide them with an ICF for their parents to sign. Additionally, every child aged 12 years or older will be invited to sign an assent form on their own behalf. The next day, upon submission of the signed ICF and assent forms, each child will receive a plastic container and be invited to produce its own urine sample. The submitted urine samples will be examined at the point-of-care (PoC) in the school using reagent strips (Haemastix; *Siemens* Healthcare Diagnostics AG; Zürich, Switzerland) and additional PoC tests if available. Haematuria-positive and/or *S. haematobium* infected children will be treated with praziquantel (40 mg/kg using a dose pole) on the day of or day after examination.

For reactive case detection in the days following the active case detection in schools, children tested positive in the schools will be asked to show a member of the study team their house of residence and the water bodies they are using for household chores or leisure activities. Household members of the infected child who are present at the time of the visit will be offered PoC testing with reagent strips (and additional PoC tests if available) and praziquantel treatment if diagnosed positive.

Moreover, the study team will visit the HWCSs indicated by the positive children, where infected individuals might have acquired the *S. haematobium* infection or introduced transmission, and implement mobile test-and-treat stalls where testing-and-treatment with reagent strips (and additional PoC tests if available) and praziquantel, respectively, will be offered to all eligible individuals using these water bodies. In addition to praziquantel treatment (40 mg/kg using a dose pole) of positive individuals at the HWCSs, response measures will include a thorough survey for the presence of intermediate host snails and molluscicide of the water bodies with niclosamide as described below.

All participants included in active and reactive surveillance will have at least the following data recorded: Sex, age, travel history, time of last praziquantel treatment, and water contact behaviour.

For passive case detection, individuals presenting with signs and symptoms of urogenital schistosomiasis in the health facilities located in the study shehias, will be tested for haematuria as a proxy for *S. haematobium* infection using reagent strips and treated with praziquantel (40 mg/kg using a dose pole) if positive. The study team and staff of the NTD programme of the MoHSWEGC will train staff of the health facilities about schistosomiasis transmission, prevention, diagnosis and treatment. Staff of the health facilities will record haematuria-positive individuals, their names and location of residency, and report them at least monthly to staff of the study team.

Intervention packages and data collection in hotspot implementation units

In the hotspot IUs, where a considerable part of the population is still infected with *S. haematobium*, we will implement a multi-disciplinary intervention package including MDA, snail control and behaviour change communication activities.

Large-scale MDA with praziquantel is the corner stone for the control and prevention of morbidity due to schistosomiasis [3, 5]. Praziquantel treatment is administered without prior diagnosis to large parts of the population living in endemic areas. In our hotspot IUs, MDA will be conducted at least annually in communities and schools by the Zanzibar NTD Programme. In communities, community drug distributors trained by the MoHSWEGC will visit all households in a door-to-door approach and offer to treat children aged 4 years or older with praziquantel (40 mg/kg) if they do not receive treatment in schools. Moreover, they will offer praziquantel to all adults that are eligible for treatment according to national treatment guidelines. In schools, praziquantel (40 mg/kg) will be provided to all children present at the day of treatment by teachers and staff of the Zanzibar NTD Programme using a dose pole [50]. The intake of tablets will be directly observed. Treatment coverage will be reported by the Zanzibar MoHSWEGC based on reports from their staff, community drug distributors, and teachers. Moreover, we will conduct our own post-treatment coverage surveys in line with the annual cross-sectional parasitology surveys in the hotspot communities and schools. When invited to submit a urine sample for annual monitoring, individuals enrolled in the survey will also be invited to respond to an ODK questionnaire, asking if they have received and swallowed the drugs provided in the MDA treatment round preceding the survey.

Snail control with the molluscicide niclsoamide is suggested by WHO as supplementary intervention to MDA in schistosomiasis-affected areas [5, 51–53]. At each identified HWCS in the hotspot IUs, a trained team will conduct regular snail surveys. While wearing wellington boots, waiders and rubber gloves to protect them from water that potentially contains infective *S. haematobium* cercariae, the team will search for snails of all species for 10 min in 20 m of the shoreline [41].

The number of snails from each collected species and data of locality, characteristics and type of water body, and observed behavioural activities will be recorded in ODK. All *Bulinus* will be taken to the laboratory for examination for *S. haematobium* infection. At all HWCSs in water bodies where *Bulinus* spp. is found, the team will apply the molluscicide niclosamide (in a concentration of 8–10 g/litre) to clear the HWCSs from infected snails and to prevent reinfection in the human population that is treated with praziquantel against schistosomiasis. Niclosamide will be sprayed to the shorelines using backpack sprayers or a petrol sprayer and the team will wear protective gear and clothing to avoid potential inhalation and eye and skin irritation [17, 54]. The amount of niclosamide used and the time and place of application will be recorded in ODK. To reduce the impact of niclosamide on other aquatic organisms, we will only focally apply the molluscicide in areas known for human water contact behaviour but not across the whole water bodies.

Behaviour change communication and community engagement is key for the success and sustainability of interventions for schistosomiasis elimination [11]. To achieve a change in behaviour, health communication, which takes local knowledge, attitudes and practices into account and integrates the communities in priority settings for decision making and planning of schistosomiasis interventions, is essential [55]. In the hotspot IUs, we will implement behavioural interventions, which were created in a human centred design approach and successfully applied in previous studies in Zanzibar [48, 56–58]. The following intervention components will be implemented: i) installation of washing platforms at a place identified together with the community in close proximity to a clean water source (i.e. pump, tap or well) to provide access and safe alternatives to washing clothes at natural water bodies; ii) training of school and madrassa teachers in classroom-based participatory teaching methods to educate children about schistosomiasis transmission and prevention using flip-charts, snail boards, blood fluke pictures, life cycle drawing, and alternative safe play methods; and iii) school-based health communication days for schistosomiasis prevention, introducing safe play methods with health education components in schools in collaboration with health teachers and classes. Within the annual cross-sectional community- and school-based surveys, we will collect KAP data including information about the perception of schistosomiasis, use of washing platforms and alternative play options, and a potential change in behaviour to assess the impact of the behaviour change and communication measures.

Laboratory procedures and data collection

The urine samples that are collected in the study will be examined with the following techniques for *S. haematobium* infections at the PoC, or in the laboratories of PHL-IdC, located in Chake Chake, Pemba.

In the annual cross-sectional parasitological surveys and for active, reactive and passive surveillance, urine samples will be screened for microhaematuria using reagent strips (Haemastix; *Siemens* Healthcare Diagnostics AG; Zürich, Switzerland). The colorimetric test results will be recorded semi-quantitatively (0 = negative, 1 = +, 2 = + +, 3 = + + +, 4 = trace).

Additionally, in the cross-sectional parasitological surveys and from a subset of samples collected during active and reactive surveillance, urine samples will be examined for the presence and number of *S. haematobium* eggs using the urine filtration method. For this purpose, urine samples will be shaken vigorously and 10 ml of each sample will be filtered through a filter-holder containing a 13 mm polycarbonate filter (Sefar, Bury, United Kingdom), using a plastic syringe [59]. All *S. haematobium* eggs present on the filter will be counted under a microscope by experienced laboratory technicians and exact egg counts will be recorded for each participant.

For external quality control, all microscope slides with the filter containing potential *S. haematobium* eggs collected during the annual cross-sectional surveys will be covered with cellophane soaked in glycerol and stored in slide storing boxes at PHL-IdC in Pemba until the end of the survey period. After each survey period, 10% of the slides will be selected based on the original electronic results by a Swiss TPH epidemiologist, sorted out locally and re-read by an external senior laboratory technician who is blinded to the results. The number of *S. haematobium* eggs counted in quality control will be recorded and compared with the original results as part of the statistical analysis done at Swiss TPH. In the case of significant discrepancies (false negatives, false positives, egg counts resulting in a different infection intensity category) in more than 20% of the re-read slides, all stored slides of the last survey will be re-read.

Both, reagent strip and urine filtration methods are not very sensitively detecting light intensity infections, as they might occur frequently in people living in elimination settings such as Pemba [29]. Hence, 10 ml from each urine samples collected at the baseline and endline survey will be stored at -20 °C and examined with a highly sensitive and specific test at the end of the study. This might be a DNA-based PCR approach [31] or an antigen-based test such as the up-converting phosphor-lateral flow circulating anodic antigen (UCP-LF) CAA assay [30] or any other test with excellent parameters developed until 2024.

To increase the performance of our surveillance-response approach, ideally a PoC test with a higher sensitivity and specificity than the reagent strips will be used. No such tests are yet (in 2021) commercially available, but several are under development. In the SchistoBreak project, we will investigate the performance of new rapid diagnostic tests, such as the Recombinase Polymerase Amplification (RPA) assay [44, 60], or other DNA-based, antigen-based or egg-based diagnostic tests for *S. haematobium* diagnosis at the PoC during our cross-sectional and surveillance activities. Once validated, applicable at the PoC, and available and affordable in sufficient numbers to screen our participants, promising candidates might be applied throughout the cross-sectional surveys and surveillance activities of the SchistoBreak project.

Participant timeline

The study will be implemented from March 2020 to June 2024, allowing for three years of interventions. Participants will be met on one or two subsequent days, for consenting, questionnaires and urine collection and examination. Figure 5 shows the timeline of surveillance-response activities in low-prevalence IUs and of the multi-pronged interventions in hotspot IUs.

Sample size

We run a series of simulations to assess how precisely the sensitivity of the surveillance system can be estimated. We modelled different scenarios with varying prevalences in schoolchildren as well as in the general population, different values for clustering and different assumptions for diagnostic test accuracy and surveillance system performance. The results revealed that sampling an average of 140 schoolchildren and 250 community members in each of 15 low-prevalence shehias in the cross-sectional surveys enables us to estimate the prevalence with a median precision (defined as one half length of the confidence interval) of 10 to 15 percentage-points in schoolchildren and of 5 to 7 percentage-points in the general population. The bias (defined as difference between the sensitivity calculated using the apparent prevalence observed during cross-sectional surveys and the sensitivity calculated using the "true" apparent prevalence observed during the surveillance activities) had a median of zero and an interquartile range from − 0.05 to 0.05.

Hence, conducting the cross-sectional surveys in the schools and communities of all 20 study shehias, the yearly total sample is 7800 individuals (ca. 10% of the population, based on an average of 4000 inhabitants per shehia).

In addition, during the surveillance activities in 15 low-prevalence shehias, we roughly estimate to test annually about 12,150 individuals (ca. 20% of the population). Among them, around 7500 will be subjected to active surveillance in high-risk schools (estimated n = 15 shehias*1 school*450 children + estimated n = 15 shehias*1 school*50 children = 7500 samples per year), 750 to reactive

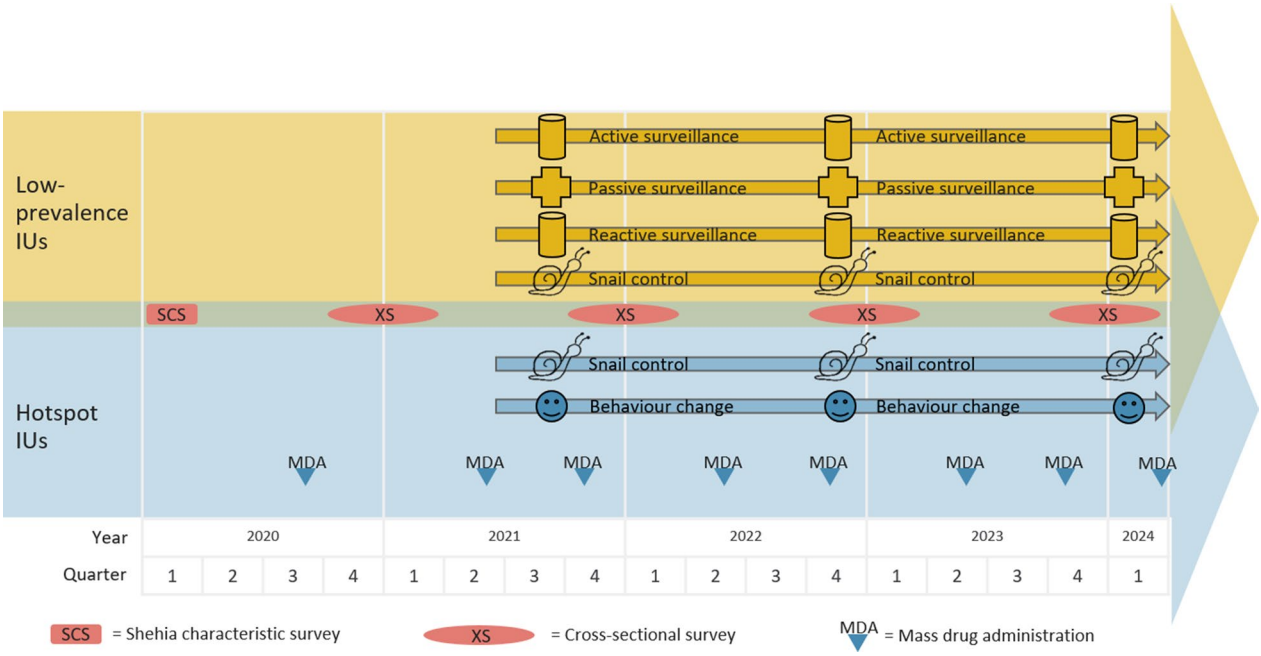


Fig. 5 Timeline of interventions against urogenital schistosomiasis in the 20 SchistoBreak study shehias in Pemba, Tanzania. IU: implementation unit

surveillance in households (estimated n = 15 shehias*10 households*5 inhabitants = 750 samples per year) and 1500 at mobile test-and-treat stalls (estimated n = 15 shehias*10 water bodies *10 people = 1500 per year), and 2400 to passive surveillance in health facilities (estimated n = 20 health facilities*12 months*10 patients = 2400 samples per year).

Data management

All data collected from individuals will include a specific personal identifier code for the individual. Laboratory data will be paper-captured before they are double entered into electronic databases by trained local data entry clerks at PHL-IdC. Double entered data will be cleaned by an epidemiologist at Swiss TPH. All discrepant electronic data will be returned to the local data entry clerks, who will trace back the personal identifier code in the original paper forms, re-enter the discrepant results and return the corrected data so that the final datasets can be generated. Registration, questionnaire and additional diagnostic data will be entered directly into the questionnaire software ODK using a tablet computer (Samsung Galaxy Tab A 2019) and subsequently uploaded on a secure server. The personal identifier code will be used to merge registration, questionnaire and laboratory examination data from each individual. Only coded data will be statistically analysed. To report the laboratory results back to the individuals, coded data can be linked with the participant names.

Data entered into electronic databases will only be accessible to authorized personnel directly involved with the study by use of a protected password. Hard copies of the source data such as student registries, laboratory record sheets, ICFs and assent forms will remain at the PHL-IdC in Pemba, respectively, for at least 10 years and stored at a safe place. The electronic versions of the data entries as well as electronic-captured data will be uploaded to and stored on a secure server from Swiss TPH.

Statistical methods

To analyse the primary outcome, we define the sensitivity of the surveillance system as the proportion of cases detected:

$$SE = N_{\text{inf detected by surveillance}}/N_{\text{inf}}$$

The total number of infected persons is unknown but will be estimated from the cross-sectional surveys as:

$$N_{\text{inf}} = P_{\text{school}} * N_{\text{school}} + P_{\text{non-school}} * N_{\text{non-school}}$$

whereby prevalences (P) and corresponding confidence intervals are estimated via generalized estimating equations for binary data with independent correlation structure to account for correlation within clusters.

The prevalence and intensity of *S. haematobium* infections will be calculated by different diagnostic approaches used in the annual cross-sectional surveys. Intensity of infection will be classified into light (< 50 eggs/10 ml urine) and heavy (≥ 50 eggs/10 ml urine) according to thresholds provided by the WHO for urine filtration results [3].

The timeliness of case notification and reactive intervention will be assessed by recording the time needed from diagnosis to treatment, from diagnosis to follow-up of *S. haematobium* positive individuals and test-and-treat of their household members, and from diagnosis to reactive snail control.

The acceptability of the surveillance-response approach will be assessed by a mixed methods approach based on the results of questionnaires run in parallel to test-and-treat approaches in low-prevalence shehias.

The coverage of test-and-treat activities will be calculated by assessing the number of people who were targeted by test-and-treat activities in relation to the total population.

MDA coverage will be calculated by assessing the number of people reporting in the cross-sectional surveys that they received praziquantel during the last MDA in relation to the total number of participants in the cross-sectional surveys.

MDA compliance in hotspot areas will be calculated by assessing the number of people reporting in the cross-sectional surveys that they have taken praziquantel as recommended, in relation to the total number of participants in the cross-sectional surveys.

The impact of snail control will be determined by assessing the snail abundance and number of intermediate host snails at each HWCS identified over time during regular snail surveys.

The impact of behaviour change interventions in schools will be evaluated by assessing knowledge, attitude and practices of schoolchildren over time based on questionnaires.

The impact of the intervention approaches on the *S. haematobium* prevalence will be analysed descriptively.

The diagnostic accuracy of new PoC tests will be calculated by determining the proportion of individuals that have been correctly identified as *S. haematobium*-infected (sensitivity) and the proportion of individuals that have been correctly identified as negative (specificity) in comparison with the reference test.

The number and location of schools, madrassas, health facilities, and water bodies and the abundance of intermediate host snails of the genus *Bulinus* identified by micro-mapping of the IUs, will be analysed descriptively.

Statistical analyses will be carried out with the statistical software STATA and R.

Discussion

In the new WHO Roadmap on NTDs published in 2021, the global elimination of schistosomiasis as a public health problem and the validated absence of infections in humans in 25 among 78 endemic countries is set as target for 2030 [5]. To achieve these goals, countries and their schistosomiasis programme managers will need clear guidance on which intervention strategies to apply, which population groups to target, which diagnostics to use, and at what thresholds to change and adapt their strategies [11–13]. In Zanzibar, which is committed to eliminate urogenital schistosomiasis in the next years, a long-term study conducted from 2012 to 2020, revealed considerable temporal and spatial heterogeneity of *S. haematobium* infections that will need to be considered in future intervention planning [6, 17, 18]. Within the SchistoBreak study we will address the focality and heterogeneity of *S. haematobium* transmission in Pemba and aim to investigate novel adaptive intervention strategies as well as standard and new diagnostic tools for schistosomiasis elimination in Zanzibar.

Working in an elimination setting, however, there are foreseeable challenges for analyses and implementation:

First, most of the population in our study area is free of schistosomiasis and there will only be very few *S. haematobium* infected individuals. Hence, even with a very large sample size involving several thousand participants, the study is underpowered and we will most likely not be able to determine the impact of our interventions in terms of statistically significant differences in the *S. haematobium* prevalence assessed in annual cross-sectional surveys. Therefore, as a computable and meaningful primary outcome, we will assess the sensitivity of the surveillance approach, based on the number of *S. haematobium* cases detected by active, reactive and passive surveillance in schools and communities divided by the total number of infected persons estimated from the cross-sectional surveys.

Second, the urine filtration and reagent strip methods that we will apply for the standard diagnosis of *S. haematobium* infection markers are not very sensitively and specifically detecting light intensity infections as primarily found in Zanzibar [29–31, 61]. Hence, in cross-sectional surveys, as well as in the surveillance-response approach, we will likely miss a considerable number of *S. haematobium*-positive individuals and underestimate the "true" prevalence. Clearly, our surveillance system will only be as good as the diagnostic methods employed. The undetected false-negative cases missed due to low test sensitivity might act as a reservoir for infections and contribute to continuous transmission. The false-positive individuals indicated by low test specificity will be chased for nothing and valuable resources and time will be wasted [13]. To improve the surveillance and get a more realistic picture of the "true" prevalence in cross-sectional surveys, in addition to the standard methods, we will apply DNA-based or antigen-based diagnostic tests, at least for the examination of urine samples at baseline and endline and, once readily available and validated, at the PoC during surveillance.

Third, to control the morbidity due to schistosomiasis, WHO provides several thresholds based on the baseline prevalence among school-aged children to decide upon the treatment strategy and frequency of MDA [3, 4]. However, to our knowledge, with regard to the schistosomiasis elimination goals, official thresholds indicating when to increase the frequency of MDA, or stop large-scale MDA and change tactics to surveillance strategies for detecting elimination or resurgence of transmission are not yet published, but urgently needed. These intervention thresholds for elimination settings should ideally be available for or transferable to a set of different diagnostic methods, which, depending on their sensitivity and specificity, will reveal different prevalences in the same population. Cross-diagnostic threshold adaptation has been evaluated for several standard tests used for the identification of both *S. haematobium* and *S. mansoni*, but only for thresholds at 10% prevalence, with samples from school-aged children, and in settings with higher intensity of infections than in Zanzibar [62, 63]. In our study, the thresholds to stratify low-prevalence IUs and hotspot IUs and the implementation of respective intervention approaches are predicated on single urine filtration results in our cross-sectional surveys and an analytical picture of the micro-epidemiology of urogenital schistosomiasis in Zanzibar established over the past 10 years. Due to age-dependent exposure rates [6, 64], we have initially selected thresholds determining hotspot IUs at $\geq 3\%$ prevalence in individuals aged ≥ 4 years sampled in community-based surveys and at $\geq 2\%$ *S. haematobium* prevalence in schoolchildren sampled in school-based surveys. The suggested thresholds will be subject to change and adaptation over the course of the project, i.e. in case we observe a considerable increase of prevalence in the low-prevalence IUs targeted with surveillance response.

Finally, good coverage will be key for the success of our intervention approaches. In the low-prevalence IUs, surveillance-response activities will start in schools and be extended in a snowball system to households of *S. haematobium*-infected individuals

and the water bodies they use. In hotspot IUs, MDA will be conducted in communities and target all eligible household members in a shehia. Schools, including not only primary schools, but also nurseries, secondary schools and madrassas, will be used as additional venues for MDA to reach as many children as possible. Snail surveys will be conducted repeatedly and multiple times per year at the HWCSs of all known water bodies in hotspot shehias. If *Bulinus* is detected in a water body, the HWCSs will be sprayed with niclosamide. Behaviour change communication including trainings in interactive teaching methods about schistosomiasis for teachers and their equipment with schistosomiasis teaching material will target all public, private and religious schools in the hotspot shehias. Public outreach days (Kichocho days) will be conducted in at least two schools per shehia and reach not only the majority of teachers and children attending the school but additional visitors from the communities. Striking health education messages and knowledge about the transmission and prevention of schistosomiasis will be transferred. Two laundry platforms per shehia will be constructed in close collaboration with the community near clean water sources to provide the communities with safe options for washing clothes. However, the implementation and coverage of the surveillance-response activities in low-prevalence IUs and the intervention package in hotspot IUs will be limited by time, costs and logistical feasibility and require careful and adaptive planning and implementation throughout the study period.

Despite the highlighted challenges, the SchistoBreak study will fill a critical health knowledge gap and produce important and much needed results. The study will show whether the performance of surveillance-response as intervention in low-prevalence IUs is sufficiently high to detect successfully all urogenital schistosomiasis cases and to respond with adequate interventions so that recrudescence of transmission can be prevented. It will also reveal if the *S. haematobium* prevalence in hotspot IUs can be reduced to very low levels when a comprehensive, multi-disciplinary intervention package is applied. Finally, the study will show whether new diagnostic tests are suitable for application at the PoC and outperforming current standard tests in their sensitivity and specificity to detect *S. haematobium* infections. Hence, our study will shed light on the field applicability and performance of novel adaptive intervention strategies and new diagnostic tools for schistosomiasis elimination and reveal whether progress towards interruption of transmission can indeed be achieved and sustained in the suggested way. The evidence and experiences generated by micro-mapping of *S. haematobium* infections at sub-district community level, micro-targeting of different intervention approaches, and application of novel diagnostic tools can guide future strategic plans for schistosomiasis elimination in Zanzibar and inform other countries aiming for interruption of transmission.

Abbreviations

GIS: Geographical Information System; HWCS: Human water contact site; ICF: Informed consent form; IU: Intervention unit; KAP: Knowledge, attitude and practice; MDA: Mass drug administration; MoHSWEGC: Ministry of Health, Social Welfare, Elderly, Gender and Children; NTD: Neglected tropical disease; PHL-IdC: Public Health Laboratory-Ivo de Carneri; PoC: Point-of-care; RPA: Recombinase Polymerase Amplification; Swiss TPH: Swiss Tropical and Public Health Institute; UCP-LF CAA: Up-converting phosphor-lateral flow circulating anodic antigen; WASH: Water, Sanitation and Hygiene; WHO: World Health Organization.

Acknowledgements

We are thankful for the support of the Zanzibar NTD Programme of the MoHSWEGC for planning and implementing the MDA, including praziquantel procurement from WHO, meetings with local authorities, and the training of drug distributors and teachers. We thank Crown Agents via the Accelerating the Sustainable Control and Elimination of Neglected Tropical Diseases (Ascend) project and the Schistosomiasis Control Initiative Foundation (SCIF) for their support with MDA implementation. We are grateful to BAYER Crop Sciences for their donation and regular quality checks of niclosamide for intermediate host snail control.

Authors' contributions

LT and SK have drafted the first version of the protocol. LT, JH, SaMA and SK conceptualized the study and SaMA and SK initiated the study. ShMA, SJ and FK advised for local conditions. SK acquired the funding for the study. LT, JH and SK conceptualized the statistical methods and JH conducted the sample size calculation of the study. All authors have made substantial contributions to the study, and have reviewed and approved the submitted version of the protocol. All authors read and approved the final manuscript.

Funding

Funding for the study has been obtained from the Swiss National Science Foundation (SNSF) via the PRIMA Grant (PR00P3_179753/1) of Stefanie Knopp. This funding source had no role in the design of this study and will not have any role during its execution, analyses, interpretation of the data, or decision to submit results.

Availability of data and materials

Not applicable.

Declarations

Ethics approval and consent to participate

The study protocol has been waived by the ethics committee of Switzerland (Ethikkommission Nordwest- und Zentralschweiz; EKNZ) on October 23, 2019 (Req-2019-00951) and has been approved by the ethics committee of Zanzibar (Zanzibar Health Research Institute; ZAHRI) on December 13, 2019 (ZAHREC/02/November/2019/16). Participants will only be included if they submit an informed consent form signed by a parent or legal guardian in case of participating children and adolescents, or signed by the participant in case of participating adults. The trial was prospectively registered at ISRCTN (ISCRCTN91431493).

Consent to publish

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author details
[1]Swiss Tropical and Public Health Institute, Socinstrasse 57, 4051 Basel, Switzerland. [2]University of Basel, Petersplatz 1, 4001 Basel, Switzerland. [3]Public Health Laboratory - Ivo de Carneri, Wawi, P.O. Box 122, Chake-Chake, Pemba, United Republic of Tanzania. [4]Neglected Diseases Program, Ministry of Health, P.O. Box 236, Zanzibar, United Republic of Tanzania.

Received: 19 August 2021 Accepted: 26 August 2021
Published online: 30 September 2021

References

1. Colley DG, Bustinduy AL, Secor WE, King CH. Human schistosomiasis. Lancet. 2014;383(9936):2253–64 (**Epub 2014/04/05**).
2. GBD 2017 DALYs and HALE Collaborators. Global, regional, and national disability-adjusted life-years (DALYs) for 359 diseases and injuries and healthy life expectancy (HALE) for 195 countries and territories, 1990–2017: A systematic analysis for the Global Burden of Disease Study 2017. Lancet. 2018;392(10159):1859–922. (**Epub 2018/11/13**).
3. WHO. Preventive chemotherapy in human helminthiasis: Coordinated use of anthelminthic drugs in control interventions: a manual for health professionals and programme managers. Geneva: World Health Organization; 2006.
4. WHO. Schistosomiasis progress report 2001–2011 and strategic plan 2012–2020. World Health Organization, 2013.
5. WHO. Ending the neglect to attain the Sustainable Development Goals—a road map for neglected tropical diseases 2021–2030. Geneva: World Health Organization; 2020.
6. Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: Repeated cross-sectional studies. PLoS Negl Trop Dis. 2021;15(2):e0009127 (**Epub 2021/02/13**).
7. Ramzy RMR, Rabiee A, Abd Elaziz KM, Campbell CH, Kittur N, Colley DG, et al. Test, treat, track, test, and treat active surveillance toward elimination of schistosomiasis: a feasibility study. Am J Trop Med Hyg. 2020;103(4):1572–7 (**Epub 2020/07/15**).
8. Shortt JA, Timm LE, Hales NR, Nikolakis ZL, Schield DR, Perry BW, et al. Population genomic analyses of schistosome parasites highlight critical challenges facing endgame elimination efforts. Sci Rep. 2021;11(1):6884.
9. Zhou Y, Chen Y, Jiang Q. History of human schistosomiasis (bilharziasis) in China: From discovery to elimination. Acta Parasitol. 2021. (**Epub 2021/03/14**).
10. Rollinson D, Knopp S, Levitz S, Stothard JR, Tchuem Tchuenté LA, Garba A, et al. Time to set the agenda for schistosomiasis elimination. Acta Trop. 2013;128(2):423–40 (**Epub 2012/05/15**).
11. Monnier N, Barth-Jaeggi T, Knopp S, Steinmann P. Core components, concepts and strategies for parasitic and vector-borne disease elimination with a focus on schistosomiasis: a landscape analysis. PLoS Negl Trop Dis. 2020;14(10):e0008837 (**Epub 2020/10/31**).
12. Tchuem-Tchuente LA, Rollinson D, Stothard JR, Molyneux D. Moving from control to elimination of schistosomiasis in sub-Saharan Africa: time to change and adapt strategies. Infect Dis Poverty. 2017;6(1):42 (**Epub 2017/02/22**).
13. Gass K. Time for a diagnostic sea-change: Rethinking neglected tropical disease diagnostics to achieve elimination. PLoS Negl Trop Dis. 2020;14(12):0008933.
14. WHO. Resolution on schistosomiasis WHA65.21 Geneva: World Health Organization; 2012. https://www.who.int/neglected_diseases/Schistosomiasis_wha65/en/.
15. Toor J, Coffeng LE, Hamley JID, Fronterre C, Prada JM, Castaño MS, et al. When, who, and how to sample: Designing practical surveillance for 7 neglected tropical diseases as we approach elimination. J Infect Dis. 2020;221(Suppl 5):S499-s502 (**Epub 2020/06/13**).
16. Knopp S, Person B, Ame SM, Mohammed KA, Ali SM, Khamis IS, et al. Elimination of schistosomiasis transmission in Zanzibar: Baseline findings before the onset of a randomized intervention trial. PLoS Negl Trop Dis. 2013;7(10):e2474 (**Epub 2013/10/23**).
17. Knopp S, Person B, Ame SM, Ali SM, Hattendorf J, Juma S, et al. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: a cluster-randomised trial. Lancet Glob Health. 2019;7(8):e1118–29 (**Epub 2019/07/01**).
18. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-Year intervention study on elimination of urogenital schistosomiasis in Zanzibar: parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019;13(5):e0007268 (**Epub 2019/05/07**).
19. Knopp S, Mohammed KA, Ali SM, Khamis IS, Ame SM, Albonico M, et al. Study and implementation of urogenital schistosomiasis elimination in Zanzibar (Unguja and Pemba islands) using an integrated multidisciplinary approach. BMC Public Health. 2012;12:930 (**Epub 2012/11/01**).
20. Stothard JR, Mgeni AF, Khamis S, Seto E, Ramsan M, Rollinson D. Urinary schistosomiasis in schoolchildren on Zanzibar Island (Unguja), Tanzania: a parasitological survey supplemented with questionnaires. Trans R Soc Trop Med Hyg. 2002;96(5):507–14.
21. WHO. Atlas of the global distribution of schistosomiasis: 29 - United Republic of Tanzania. 1987.
22. Mott KE. Contrasts in the control of schistosomiasis. Mem Inst Oswaldo Cruz. 1989;84(Suppl 1):3–19 (**Epub 1989/10/01**).
23. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba island: programme organization and management. Trop Med Parasitol. 1989;40(2):189–94.
24. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba island; selective population chemotherapy of schoolchildren with haematuria to identify high-risk localities. Trans R Soc Trop Med Hyg. 1989;83(6):805–10 (**Epub 1989/11/01**).
25. Albonico M, Chwaya HM, Montresor A, Stolfzfus RJ, Tielsch JM, Alawi KS, et al. Parasitic infections in Pemba Island school children. East Afr Med J. 1997;74(5):294–8 (**Epub 1997/05/01**).
26. Mgeni AF, Kisumku UM, McCullough FS, Dixon H, Yoon SS, Mott KE. Metrifonate in the control of urinary schistosomiasis in Zanzibar. Bull World Health Organ. 1990;68(6):721–30.
27. Stothard JR, Rollinson D. Molecular characterization of *Bulinus globosus* and *B. nasutus* on Zanzibar, and an investigation of their roles in the epidemiology of *Schistosoma haematobium*. Trans R Soc Trop Med Hyg. 1997;91(3):353–7 (**Epub 1997/05/01**).
28. Rollinson D, Stothard JR, Southgate VR. Interactions between intermediate snail hosts of the genus *Bulinus* and schistosomes of the *Schistosoma haematobium* group. Parasitology. 2001;123(Suppl):S245–60 (**Epub 2002/01/05**).
29. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Urogenital schistosomiasis elimination in Zanzibar: accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections. Parasit Vectors. 2018;11(1):552 (**Epub 2018/10/26**).
30. Knopp S, Corstjens PL, Koukounari A, Cercamondi CI, Ame SM, Ali SM, et al. Sensitivity and specificity of a urine circulating anodic antigen test for the diagnosis of *Schistosoma haematobium* in low endemic settings. PLoS Negl Trop Dis. 2015;9(5):e0003752 (**Epub 2015/05/15**).
31. Keller D, Rothen J, Dangy JP, Saner C, Daubenberger C, Allan F, et al. Performance of a real-time PCR approach for diagnosing *Schistosoma haematobium* infections of different intensity in urine samples from Zanzibar. Infect Dis Poverty. 2020;9(1):128–30 (**Epub 2020/09/06**).
32. Colley DG, Fleming FM, Matendechero SH, Knopp S, Rollinson D, Utzinger J, et al. Contributions of the Schistosomiasis Consortium for Operational Research and Evaluation (SCORE) to schistosomiasis control and elimination: Key findings and messages for future goals, thresholds, and operational research. Am J Trop Med Hyg. 2020;103(1_Suppl):125–34. Epub 2020/05/14.
33. Gardner JMF, Mansour NR, Bell AS, Helmby H, Bickle Q. The discovery of a novel series of compounds with single-dose efficacy against juvenile and adult *Schistosoma* species. PLoS Negl Trop Dis. 2021;15(7):e0009490-91 (**Epub 2021/07/20**).
34. Bergquist R, Yang GJ, Knopp S, Utzinger J, Tanner M. Surveillance and response: tools and approaches for the elimination stage of neglected tropical diseases. Acta Trop. 2015;141(Pt B):229–34 (**Epub 2014/10/11**).
35. Qian MB, Chen J, Bergquist R, Li ZJ, Li SZ, Xiao N, et al. Neglected tropical diseases in the People's Republic of China: Progress towards elimination. Infect Dis Poverty. 2019;8(1):86 (**Epub 2019/10/04**).
36. Liang S, Yang C, Zhong B, Guo J, Li H, Carlton EJ, et al. Surveillance systems for neglected tropical diseases: Global lessons from China's evolving schistosomiasis reporting systems, 1949–2014. Emerg Themes Epidemiol. 2014;11:19 (**Epub 2014/01/01**).

37. Bezerra DVF, Queiroz JW, Câmara VAV, Maciel BLL, Nascimento ELT, Jerônimo SMB. Factors associated with *Schistosoma mansoni* infestation in northeast Brazil: A need to revisit individual and community risk factors. Am J Trop Med Hyg. 2021. Epub 2021/02/17.
38. Sokolow SH, Wood CL, Jones IJ, Swartz SJ, Lopez M, Hsieh MH, et al. Global assessment of schistosomiasis control over the past century shows targeting the snail intermediate host works best. PLoS Negl Trop Dis. 2016;10(7):e0004794 (**Epub 2016/07/22**).
39. Ng'etich AKS, Voyi K, Mutero CM. Assessment of surveillance core and support functions regarding neglected tropical diseases in Kenya. BMC Public Health. 2021;21(1):142 (**Epub 2021/01/17**).
40. Rudge JW, Stothard JR, Basanez MG, Mgeni AF, Khamis IS, Khamis AN, et al. Micro-epidemiology of urinary schistosomiasis in Zanzibar: Local risk factors associated with distribution of infections among school-children and relevance for control. Acta Trop. 2008;105(1):45–54 (**Epub 2007/11/13**).
41. Pennance T, Person B, Muhsin MA, Khamis AN, Muhsin J, Khamis IS, et al. Urogenital schistosomiasis transmission on Unguja Island, Zanzibar: Characterisation of persistent hot-spots. Parasit Vectors. 2016;9(1):646–8 (**Epub 2016/12/18**).
42. Souza AA, Ducker C, Argaw D, King JD, Solomon AW, Biamonte MA, et al. Diagnostics and the neglected tropical diseases roadmap: setting the agenda for 2030. Trans R Soc Trop Med Hyg. 2021;115(2):129–35 (**Epub 2020/11/11**).
43. Mabey D, Agler E, Amuasi JH, Hernandez L, Hollingsworth TD, Hotez PJ, et al. Towards a comprehensive research and development plan to support the control, elimination and eradication of neglected tropical diseases. Trans R Soc Trop Med Hyg. 2021;115(2):196–9.
44. Archer J, Barksby R, Pennance T, Rostron P, Bakar F, Knopp S, et al. Analytical and clinical assessment of a portable, isothermal recombinase polymerase amplification (RPA) assay for the molecular diagnosis of urogenital schistosomiasis. Molecules. 2020;25:18 (**Epub 2020/09/17**).
45. WHO. Communicable disease surveillance and response systems. Guide to monitoring and evaluation. Geneva: World Health Organization; 2006.
46. Office of Chief Government Statistician Zanzibar. Zanzibar in figures 2019. 2020.
47. Office of Chief Government Statistician Zanzibar. 2012 Population and housing census: Population distribution by admistrative areas. 2013.
48. Celone M, Person B, Ali SM, Lyimo JH, Mohammed UA, Khamis AN, et al. Increasing the reach: Involving local Muslim religious teachers in a behavioral intervention to eliminate urogenital schistosomiasis in Zanzibar. Acta Trop. 2016;163:142–8 (**Epub 2016/08/09**).
49. Office of Chief Government Statistician Zanzibar. Household budget survey 2014/2015. 2016.
50. Montresor A, Engels D, Ramsan M, Foum A, Savioli L. Field test of the "dose pole" for praziquantel in Zanzibar. Trans R Soc Trop Med Hyg. 2002;96(3):323–4 (**Epub 2002/08/15**).
51. King CH, Sutherland LJ, Bertsch D. Systematic review and meta-analysis of the impact of chemical-based mollusciciding for control of *Schistosoma mansoni* and *S. haematobium* transmission. PLoS Negl Trop Dis. 2015;9(12):0004290.
52. WHO. The Role of mollusciciding in schistosomiasis control. Geneva: World Health Organization; 1992.
53. Sturrock RF. Complementary snail control in chemotherapeutic-based control programs. In: Miller MJL, E., editor. Parasitic Diseases: Treatment & Control, 1989. p. 51–7.
54. Allan F, Ame SM, Tian-Bi YT, Hofkin BV, Webster BL, Diakité NR, et al. Snail-related contributions from the Schistosomiasis Consortium for Operational Research and Evaluation program including xenomonitoring, focal molluscicide, biological control, and modeling. Am J Trop Med Hyg. 2020;103(1_Suppl):66–79. Epub 2020/05/14.
55. Aagaard-Hansen J, Mwanga JR, Bruun B. Social science perspectives on schistosomiasis control in Africa: past trends and future directions. Parasitology. 2009;136(13):1747–58.
56. Person B, Knopp S, Ali SM, A'kadir FM, Khamis AN, Ali JN, et al. Community co-designed schistosomiasis control interventions for school-aged children in Zanzibar. Journal of Biosocial Science. 2016;48(S1):S56-S73. Epub 2016/07/18.
57. Person B, Ali SM, A'Kadir FM, Ali JN, Mohammed UA, Mohammed KA, et al. Community knowledge, perceptions, and practices associated with urogenital schistosomiasis among school-aged children in Zanzibar, United Republic of Tanzania. PLoS Negl Trop Dis. 2016;10(7):e0004814 (**Epub 2016/07/12**).
58. IDEO.org. The field guide to human-centered design: IDEO.org; 2015. 192 p.
59. Peters PA, Mahmoud AA, Warren KS, Ouma JH, Siongok TK. Field studies of a rapid, accurate means of quantifying *Schistosoma haematobium* eggs in urine samples. Bull World Health Organ. 1976;54(2):159–62 (**Epub 1976/01/01**).
60. Rostron P, Pennance T, Bakar F, Rollinson D, Knopp S, Allan F, et al. Development of a recombinase polymerase amplification (RPA) fluorescence assay for the detection of *Schistosoma haematobium*. Parasit Vectors. 2019;12(1):514.
61. Krauth SJ, Greter H, Stete K, Coulibaly JT, Traoré SI, Ngandolo BN, et al. All that is blood is not schistosomiasis: Experiences with reagent strip testing for urogenital schistosomiasis with special consideration to very-low prevalence settings. Parasit Vectors. 2015;8:584 (**Epub 2015/11/12**).
62. Midzi N, Bärenbold O, Manangazira P, Phiri I, Mutsaka-Makuvaza MJ, Mhlanga G, et al. Accuracy of different diagnostic techniques for *Schistosoma haematobium* to estimate treatment needs in Zimbabwe: application of a hierarchical Bayesian egg count model. PLoS Negl Trop Dis. 2020;14(8):e0008451 (**Epub 2020/08/21**).
63. Bärenbold O, Garba A, Colley DG, Fleming FM, Haggag AA, Ramzy RMR, et al. Translating preventive chemotherapy prevalence thresholds for *Schistosoma mansoni* from the Kato-Katz technique into the point-of-care circulating cathodic antigen diagnostic test. PLoS Negl Trop Dis. 2018;12(12):0006941 (**Epub 2018/12/15**).
64. Toor J, Rollinson D, Turner HC, Gouvras A, King CH, Medley GF, et al. Achieving elimination as a public health problem for *Schistosoma mansoni* and *S. haematobium*: when is community-wide treatment required? J Infect Dis. 2020;221(Suppl 5):S525–30.

Publisher's Note

3.4. GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation

Lydia Trippler[1,2], Mohammed Nassor Ali[3], Shaali Makame Ame[3], Said Mohammed Ali[3], Fatma Kabole[4], Jan Hattendorf[1,2], Stefanie Knopp[1,2]

1 Swiss Tropical and Public Health Institute, Allschwil, Switzerland

2 University of Basel, Basel, Switzerland

3 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

4 Neglected Diseases Programme, Zanzibar Ministry of Health, Social Welfare, Elderly, Gender and Children, Lumumba, Unguja, United Republic of Tanzania

This article was published in:

BMC Infectious Diseases of Poverty (2022), Jan 15; 11(1):8

doi: 10.1186/s40249-021-00928-y

Infectious Diseases of Poverty

RESEARCH ARTICLE Open Access

GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation

Lydia Trippler[1,2], Mohammed Nassor Ali[3], Shaali Makame Ame[3], Said Mohammed Ali[3], Fatma Kabole[4], Jan Hattendorf[1,2] and Stefanie Knopp[1,2*]

Abstract

Background: Fine-scale mapping of schistosomiasis to guide micro-targeting of interventions will gain importance in elimination settings, where the heterogeneity of transmission is often pronounced. Novel mobile applications offer new opportunities for disease mapping. We provide a practical introduction and documentation of the strengths and shortcomings of GPS-based household identification and participant recruitment using tablet-based applications for fine-scale schistosomiasis mapping at sub-district level in a remote area in Pemba, Tanzania.

Methods: A community-based household survey for urogenital schistosomiasis assessment was conducted from November 2020 until February 2021 in 20 small administrative areas in Pemba. For the survey, 1400 housing structures were prospectively and randomly selected from shapefile data. To identify pre-selected structures and collect survey-related data, field enumerators searched for the houses' geolocation using the mobile applications Open Data Kit (ODK) and MAPS.ME. The number of inhabited and uninhabited structures, the median distance between the pre-selected and recorded locations, and the dropout rates due to non-participation or non-submission of urine samples of sufficient volume for schistosomiasis testing was assessed.

Results: Among the 1400 randomly selected housing structures, 1396 (99.7%) were identified by the enumerators. The median distance between the pre-selected and recorded structures was 5.4 m. A total of 1098 (78.7%) were residential houses. Among them, 99 (9.0%) were dropped due to continuous absence of residents and 40 (3.6%) households refused to participate. In 797 (83.1%) among the 959 participating households, all eligible household members or all but one provided a urine sample of sufficient volume.

Conclusions: The fine-scale mapping approach using a combination of ODK and an offline navigation application installed on tablet computers allows a very precise identification of housing structures. Dropouts due to non-residential housing structures, absence, non-participation and lack of urine need to be considered in survey designs. Our findings can guide the planning and implementation of future household-based mapping or longitudinal surveys and thus support micro-targeting and follow-up of interventions for schistosomiasis control and elimination in remote areas.

*Correspondence: s.knopp@swisstph.ch
[1] Swiss Tropical and Public Health Institute, Kreuzstrasse 2, 4123 Allschwil, Switzerland
Full list of author information is available at the end of the article

Trial registration ISRCTN, ISCRCTN91431493. Registered 11 February 2020, https://www.isrctn.com/ISRCTN91431493

Keywords: Urogenital schistosomiasis, Control, Elimination, Intervention, Fine-scale mapping, Interruption of transmission, Micro-mapping, Precision mapping, Wayfinding, Zanzibar

Background

Schistosomiasis is among the 20 neglected tropical diseases (NTDs) defined by the World Health Organization (WHO) and is endemic in 78 countries worldwide [1–3]. The causative agent of schistosomiasis is a blood fluke of the genus *Schistosoma* that infects more than 200 million people worldwide [4]. The parasite is transmitted through skin contact with freshwater containing the infectious larval stages, which are released by an aquatic intermediate host snail vector [5]. Over the past decades, great progress was made in schistosomiasis control [6, 7]. In their new roadmap for neglected tropical diseases 2021–2030, the WHO set the global elimination of schistosomiasis as a public health problem and the validated absence of infection in humans in 25 among the 78 endemic countries as targets for 2030 [1].

The sustained implementation of control interventions, including mass drug administration (MDA) as the cornerstone, often supplemented with snail control, educational measures, or improvements in the socio-economic standard and access to clean water and sanitation, has resulted in decreasing schistosomiasis prevalences in many countries [8–11]. In areas where prevalences decline, the focality and heterogeneity of schistosomiasis becomes more pronounced [8, 12–14]. Typically, within a few years of annual or biannual MDA across districts and countries, many communities reach low schistosomiasis prevalence and infection intensity [10, 15, 16]. Some areas, however, remain as pockets of high transmission with persistent or reoccurring high prevalence levels despite intense interventions [8, 12–14, 17–20].

To achieve the WHO elimination goals, the pronounced spatial and temporal heterogeneity in schistosomiasis elimination settings will need to be considered in future intervention planning [8, 14, 20–23].

Fine-scale mapping of schistosomiasis is considered an essential requirement to move from morbidity control towards interruption of transmission in endemic areas [23]. Detailed fine-scale mapping of infection and disease patterns can help to identify low-risk and high-risk areas of transmission at sub-district level and to micro-target interventions in line with infection levels for optimal treatment and resource allocation [10, 14, 24, 25]. Android tablet-based applications containing or being able to connect to high-resolution maps to geolocate individuals' residences in remote resource poor areas have been suggested as attractive approaches for fine-scale disease mapping [26, 27].

Here, we provide a practical introduction and documentation of the strengths and shortcomings of Global Position System (GPS)-based household identification and participant recruitment using Android tablet-based applications for fine-scale schistosomiasis mapping in a remote area in Pemba, United Republic of Tanzania.

Methods

Study area

The fine-scale mapping approach described here is part of the cross-sectional baseline survey of the "SchistoBreak" study, a multi-year project that aims to investigate new tools and strategies for breaking schistosomiasis transmission [28]. Pemba and Unguja are the main islands of the Zanzibar Archipelago, a semi-autonomous part of the United Republic of Tanzania. Pemba, where our study is implemented, is divided into four districts: Micheweni, Wete, Chake Chake and Mkoani [29]. In total, the districts in Pemba contain 129 small administrative areas, which are called shehias [30]. The study area of the SchistoBreak project, where the here reported fine-scale mapping survey is part of, covers a total of 20 shehias in the north of Pemba. Among them, 12 shehias are located in Micheweni and eight in Wete district, respectively. The average population size in our study shehias was ~3900 individuals per shehia in 2012, with a minimum of ~2200 and a maximum of ~8600 inhabitants [31]. As indicated in Fig. 1, shehias in Pemba are mostly connected by tarmacked main roads. However, some rough road connections exist. Housing structures within the shehias are grouped in clusters, which are usually aligned along the main roads or located near unpaved streets or footpaths. Housing structures are mostly not characterized or identifiable by street names or house numbers. Urogenital schistosomiasis is endemic in the north of Pemba [32, 33]. Due to intense control and elimination efforts implemented across the Zanzibar islands over the past decades, the overall *S. haematobium* prevalence had reached a very low level of 3.4% in schoolchildren in 2020, constituted by many low-prevalence areas and a few remaining hotspot areas of transmission [8].

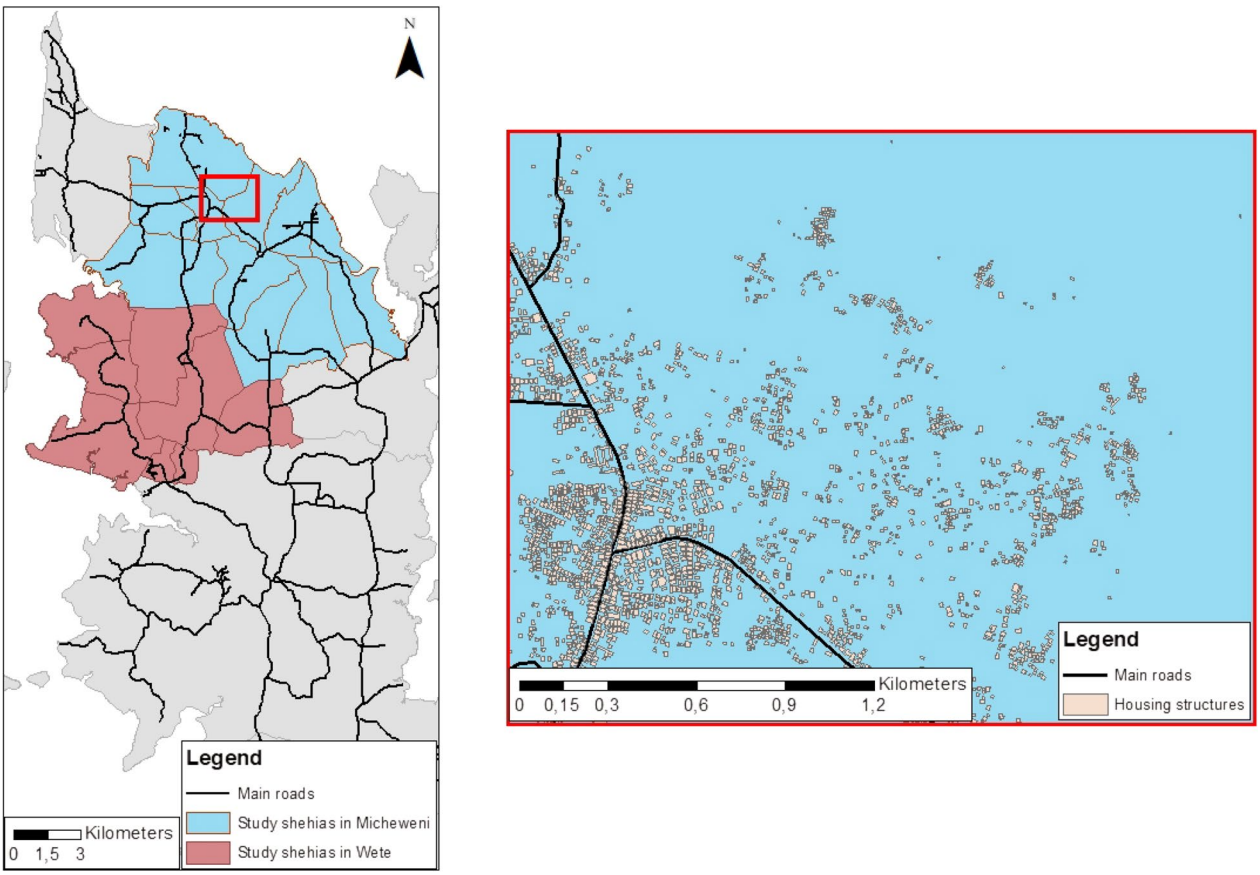


Fig. 1 Study area of the SchistoBreak project in Pemba, Tanzania. The exemplary section shows the distribution of housing structures and main roads. The image base map (United Republic of Tanzania—Subnational administrative boundaries) was downloaded from OCHA services (https://data.humdata.org/dataset/tanzania-administrative-boundaries-level-1-to-3-regions-districts-and-wards-with-2012-population). The data source is: Tanzania National Bureau of Statistics/UN OCHA ROSA. The data are published under the following license: Creative Commons Attribution for Intergovernmental Organisations [CC BY-IGO; (https://creativecommons.org/licenses/by/3.0/igo/legalcode)]. Additionally, we received written permission to use and adapt the data from the UN Office for the Coordination of Humanitarian Affairs (OCHA). Additional shapefiles for the map (shehia boundaries and housing structures) were provided by the Zanzibar Commission for Lands to the Zanzibar Neglected Diseases Program

Design of cross-sectional household survey

In the baseline survey of the SchistoBreak study, we aimed to assess the apparent prevalence of *S. haematobium* infections in the 20 study shehias in the north of Pemba. To gain a clear picture of the schistosomiasis distribution we conducted a cross-sectional community-based household survey from November 2020 until February 2021, based on randomly selected housing structures. For fine-scale mapping, we aimed to include 50 households with an average of five household members per shehia, and hence a total of 250 individuals per shehia. Details of the sample size calculations are provided in our published study protocol [28]. Eligible to participate in the household survey were all individuals aged 4 years or older living in the study shehias. Accounting for an estimated 30% dropout due to housing structures not being residential houses or refusal of household members to participate in the study, we randomized 70 housing structures per shehia. Our survey team consisted of seven field-enumerators. Each of them visited 10 housing structures per shehia. Shehias were visited for 3 subsequent days to allow enough time for revisiting houses in case the inhabitants were absent on Day 1 and/or for delayed urine submission on Day 2 or Day 3. Each enumerator was equipped with a tablet computer for navigation to their pre-selected housing structures using the offline navigation map application © 2020 MAPS.ME (https://www.maps.me/) and for electronic capturing of participant data using the data collection software © 2020 Open Data Kit (ODK; https://www.opendatakit.org/).

Randomization of housing structures

For the randomization of houses, we used shapefile data of housing structures that were provided by the Zanzibar Commission for Lands to the Zanzibar Neglected Diseases Program. Shape file data could have also been obtained from © OpenStreetMap contributors (OSM). The OSM house shapefile data are published under the Open Database License (https://www.openstreetmap.org/copyright). Housing structures are indicated as polygons in shapefiles. For randomization of housing structures, for navigating to the randomized structures, and for conducting the questionnaire, we used the statistical software R version 3.5.1 (https://www.r-project.org/), the geospatial analysis software Esri ArcMap version 10.6.1, the offline map application MAPS.ME, and the data collection software ODK, respectively.

For randomization of the 70 housing structures, we first imported the shapefile polygon data into ArcMap and used its feature-to-point tool to calculate the centroid points of all housing polygons. As the information about the location of the centroids was provided in meters, we converted the meters into latitude and longitude coordinates as this is required by MAPS.ME. Subsequently, we exported the centroids given as shapefile data from ArcMap into R and randomly selected 70 houses per shehia.

Stratification of housing structures per shehia and enumerator

After randomly selecting 70 houses per shehia in R, we imported the centroids of randomized housing structures back into ArcMap. With the selection-by-lasso tool of ArcMap, we stratified the centroids in equal-sized groups of 10 per each of the seven enumerators. Hereby, we took care that the 10 housing structures for each enumerator were as close in distance as possible to avoid the need for far-distance walking. We saved each group of housing structures per enumerator as a separate layer and shapefile and subsequently exported all single shapefiles to R. In R, we added a new variable "interviewer" and assigned a unique value to each group of housing structures per enumerator. With the new interviewer variable, we ensured to keep the group of housing structures in the next step, in which we combined all single shapefiles of housing groups into one large dataset. Subsequently, we saved the merged data containing the shehia name, the geolocation of all 70 randomized housing structures per shehia, and the group variable for seven groups with 10 housing structures each per shehia as csv-files and as shapefiles for display.

Import of grouped housing structures per enumerator into ODK

Once the randomization process was completed, each housing structure was given a unique identifier code (with the variable name: "houseID"), which together with the shehia name, and latitude and longitude coordinates, was exported from the csv-files into an ODK excel sheet. In order to limit the choices displayed in ODK Collect to only those that belonged to the selected enumerator's group of housing structures, we used the "choice_filter" option of ODK. An example for using the choice_filter option with regards to the shehia and the enumerator is presented in Table 1, line 1–3 and Table 2, line 1–12. The example in Tables 1 and 2 shows a model for two shehias and two enumerators, each taking care of one group of housing structures per shehia.

Combination of MAPS.ME and ODK for wayfinding to selected housing structures

The enumerators used the navigator app MAPS.ME to identify and find the way to each randomized housing structure in each shehia. We selected this app as it works offline and downloadable maps contain the housing structures and street data published on OSM. ODK and MAPS.ME apps were installed on the mobile devices (Samsung Galaxy Tab A tablets; Samsung Electronics, Seoul, South Korea) that were used by the enumerators. To connect ODK with MAPS.ME, we inserted into the ODK excel sheet the information presented in Table 1, line 3–8, and Table 2, line 5–15. As indicated in Table 1, line 3–5, the calculation of the geolocation is based on the houseID that is selected by the enumerator and presented in Table 2, line 5–12. Table 1, line 6 and the corresponding lines 13–15 in Table 2 serve to select different transport modes, e.g. walking, going by vehicle or by bicycle so that transport ways and transport time can be calculated by MAPS.ME. By opening the ODK questionnaire, choosing a houseID and selecting the transport mode on the terminal mobile device, a button saying "Click to open maps.me" appears as indicated in Table 1, line 7. By clicking the appearing button, the prior installed navigator app MAPS.ME opens and the geolocation of the selected houseID is automatically set as destination point and displays the way to go from the point of being to the housing structure of interest.

Community-based data collection

To allow enough time for household identification, participant recruitment, questionnaire interview and urine collection, a shehia was visited by the field-enumerators for three subsequent days. On Day 1, most or all of the 10 pre-selected housing structures per enumerator were

Table 1 Customizing the ODK excel sheet for data collection and combined use with MAPS.ME: the survey sheet

line	type	name	label::english (en)	appearance	calculation	choice_filter
1	select_one interviewer	interviewer	Interviewer:			
2	select_one shehia	shehia	Shehia:			
3	select_one houseID	houseID	To which household are you going?			shehia = ${shehia} and interviewer = ${interviewer}
4	calculate	latitude			instance('houseID')/root/item[name = ${houseID}]/latitude	
5	calculate	longitude			instance('houseID')/root/item[name = ${houseID}]/longitude	
6	select_one modes	mode	How will you get there?			
7	integer	mapsme-wayfinding	Click to open maps.me	ex:com.mapswithme.maps.pro.action BUILD_ROUTE(lat_to = number(${latitude}), lon_to = number(${longitude}), router = ${mode})		
8	select_one modes	goodbye	Thanks!			latitude = 1 and longitude = 1

Table 2 Customizing the ODK excel sheet for data collection and combined use with MAPS.ME: the choices sheet

line	list_name	name	label::english (en)	shehia	interviewer	latitude	longitude
1	interviewer	interviewer 1	interviewer 1				
2	interviewer	interviewer 2	interviewer 2				
3	shehia	shehia 1	Shehia 1				
4	shehia	shehia 2	Shehia 2				
5	houseID	houseID-01	HouseID-01	shehia 1	interviewer 1	*.***** [a]	**.*****
6	houseID	houseID-02	HouseID-02	shehia 1	interviewer 1	*.*****	**.*****
7	houseID	houseID-03	HouseID-03	shehia 1	interviewer 2	*.*****	**.*****
8	houseID	houseID-04	HouseID-04	shehia 1	interviewer 2	*.*****	**.*****
9	houseID	houseID-05	HouseID-05	shehia 2	interviewer 1	*.*****	**.*****
10	houseID	houseID-06	HouseID-06	shehia 2	interviewer 1	*.*****	**.*****
11	houseID	houseID-07	HouseID-07	shehia 2	interviewer 2	*.*****	**.*****
12	houseID	houseID-08	HouseID-08	shehia 2	interviewer 2	*.*****	**.*****
13	modes	pedestrian	Walking				
14	modes	vehicle	By bus or car				
15	modes	bicycle	By bicycle				

[a] Geolocations are not shown to preserve confidentiality

identified in the shehia community, and the geolocation and type of housing structure was recorded. In case the house was inhabited, household members were invited to participate in the study. Once a present adult household member agreed to participate, a questionnaire interview was conducted and instructions for urine collection from all household members aged ≥ 4 years were provided. One urine collection container per eligible household member was distributed and labelled with a unique picture (e.g. with a boat, star, cat or smiley) linked to the name of the participant recorded on a paper sheet to avoid confusion of urine samples. On Day

2, written informed consent forms for each participant, either signed by the participant or, in case of children, by their parent or legal guardian, were collected by the enumerator together with the urine samples from the participants. For all children 12–17 years old, an additional assent form signed by the adolescents themselves was also collected. Day 3 served as a mop-up day, when urine samples that were not submitted on Day 1 or Day 2 were collected.

Data management and statistical analysis

All data were collected with ODK and transferred to the ODK Central Server hosted at Swiss Tropical and Public Health Institute (Swiss TPH). All data were cleaned and analyzed with R version 3.5.1. Anonymized data are available as Additional files 1, 2. To document the strengths and shortcomings of GPS-based household identification and participant recruitment for fine-scale schistosomiasis mapping in a remote area in Africa (and in our case Pemba island), we evaluated the results of the community-based household survey once the data collection was completed.

First, we calculated the number of identified housing structures that were no residential houses but other structures such as buildings under construction, schools or shops.

Second, to assess the concordance of the location of the pre-selected housing structure with the location of the housing structures identified in the shehia communities by the enumerator, we determined the median, minimum and maximum distance between the initially randomized centroids and the recorded geolocations across all shehias and per shehia.

Third, we determined the number of residential houses where inhabitants were not at home at any day of the visits or where all inhabitants refused to participate in the study. The dropout rate of housing structures was calculated by dividing the number of not included housing structures by the total number of initially selected housing structures.

Fourth, for households that agreed to participate in the study, we assessed the total number of household members per inhabited house and the number of individuals eligible to participate in the study. The dropout rate of individuals who were eligible but did not provide a urine sample of sufficient volume for schistosomiasis testing was calculated by dividing the number of participants who submitted a urine sample by the total number of eligible individuals, stratified by sex and age category (i.e. adults versus children, defined as individuals below the age of 18 years).

Finally, we determined the number of urine samples received from each household and calculated the percentage of households where all participants provided a urine sample of sufficient volume for testing.

Results

Housing structures and participating households

Among the 1400 housing structures that were initially randomly selected to be surveyed in the 20 shehias, the enumerators were able to locate and identify 1396 housing structures (99.7%) (Fig. 2). Four housing structures were not identified due to inaccessibility. Among the 1396 housing structures correctly identified, 298 (21.3%) were no residential houses: 99 were buildings under constructions, 57 were broken housing structures, 22 were Islamic schools, 21 were mosques, 18 were shops, 15 were animal stalls and 66 were other buildings, such as public schools and offices. Among the 1098 residential houses, in 99 (9.0%) houses, inhabitants were absent during any of the visits of the enumerator and in 40 (3.6%) houses, inhabitants did not agree to participate in the household survey. Hence, the overall dropout rate of housing structures was 31.5% (441/1400).

Distance between centroids and recorded geolocation of housing structures

As indicated in Fig. 3, the median distance between the randomized centroids of the housing structures and the geolocation recorded by the enumerator when arriving at the housing structure was 5.4 m (range: 0.2–940.3 m). While in the six shehias surveyed in the early study period (weeks 1–4) the median distance was 7.4 m (range: 0.3–940.3 m), in the 14 shehias surveyed in the later study period (week 4–12) the median distance was 4.8 m (range: 0.2–54.4 m).

Among the 1396 identified housing structures, four recorded geolocations were more than 100 m away from the pre-selected geopoints, taken by four different enumerators. In general, for all seven enumerators there was no major difference in the distribution of distances between the preselected and recorded geopoints (Fig. 4).

Study participants

In total, 5340 individuals aged 0–99 years lived in the 959 participating households. In average, one household consisted of six individuals (range: 1–16 inhabitants).

Among the 5340 individuals, 4599 (86.1%) were eligible to participate in the study since their age was ≥ 4 years. Among them, 2324 (50.5%) were female and 2275 (49.5%) were male, and 2364 (51.4%) individuals were adults aged ≥ 18 years and 2233 (48.6%) were

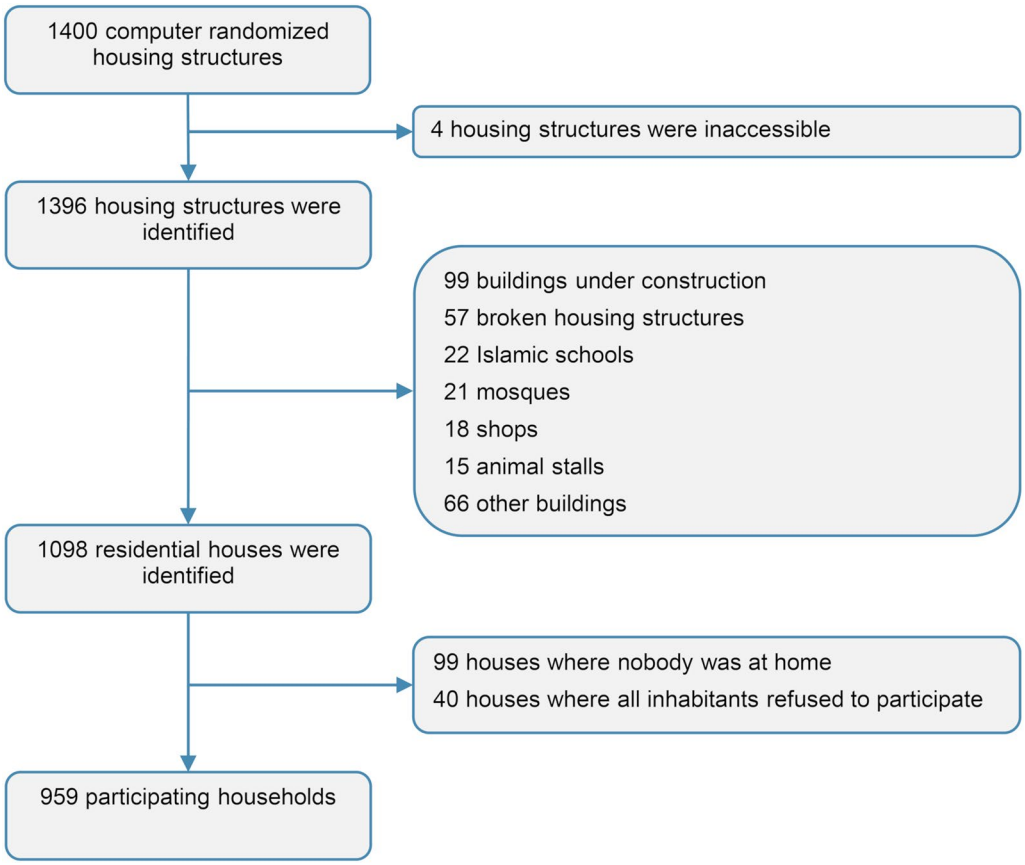


Fig. 2 Housing structures and households participating in the fine-scale mapping survey in Pemba, Tanzania

children aged 4–17 years. Two participants did not report their age. A total of 713 participants did not provide a urine sample, resulting in a dropout rate of 15.5% (713/4599). The dropout rate was higher in males (19.8%, 450/2275) than females (11.3%, 263/2323) and in adults (18.6%, 440/2364) compared with children (12.2%, 272/2233).

The number of participating households per shehia ranged from 40 to 56. In six among the 20 study shehias, the number of 50 households we aimed to include was achieved or exceeded (Fig. 5A). The number of participating individuals per shehia ranged from 171 to 319. In five shehias, the number of 250 participants we aimed for was achieved or exceeded (Fig. 5B).

Among the 959 participating households, in 598 (62.4%) households all eligible members provided a urine sample of sufficient volume ($\geq$ 10 ml) for laboratory examinations, in 199 (20.8%) households all inhabitants but one individual provided a sufficiently large urine sample, in 81 (8.4%) households all inhabitants but two provided a sufficiently large urine sample, and in 81 (8.4%) households all inhabitants but three or more individuals provided a sufficiently large urine sample (Fig. 6).

Discussion

In areas where schistosomiasis control progresses towards elimination, heterogeneity of transmission becomes more pronounced and fine-scale mapping of infection patterns is needed for effective micro-targeting of interventions at sub-district level [14, 23, 24, 28]. Novel tablet-based tools containing offline high-resolution maps offer new opportunities for disease mapping and household identification, particularly in remote areas where the distribution of housing structures do not follow an order such as street names or house numbers and where internet connectivity is poor [26].

Here, we aimed to provide a practical introduction and documentation of the strengths and shortcomings of tablet application-based household identification and participant recruitment for fine-scale schistosomiasis mapping in a rural area in Pemba, United Republic of Tanzania.

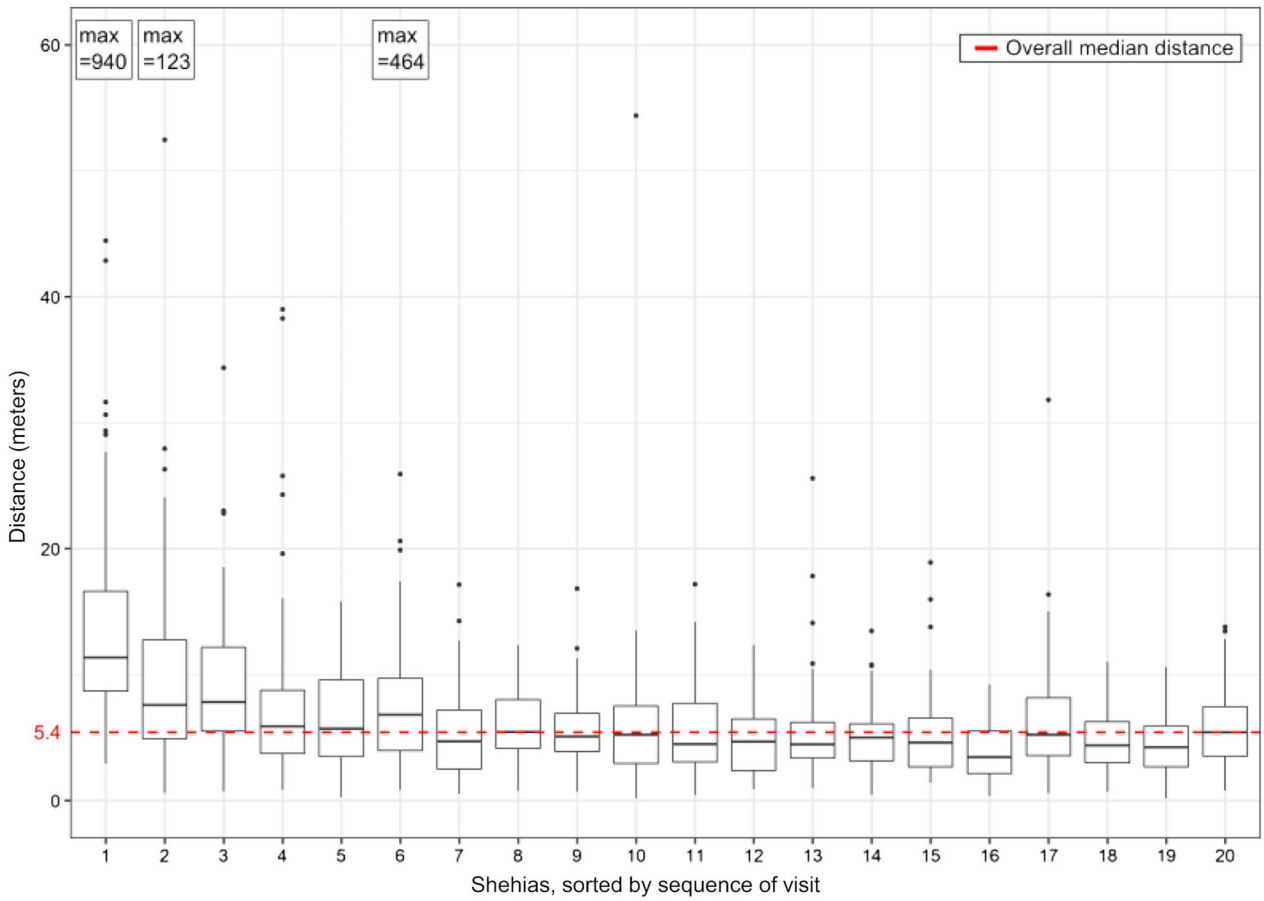


Fig. 3 Distance between pre-selected and recorded geolocations of housing structures in the mapping survey in Pemba, Tanzania, per shehia. The horizontal middle lines in each boxplot indicate the median distance per shehia, the boxes indicate the lower and upper quartile, the vertical lines indicate the minimum and maximum excluding outliers, and the dots indicate outliers (more than 1.5 times the interquartile range below the first or above the 3rd quartile). For shehia 1, 2 and 6 the maximum outliers are indicated separately. The red dashed line indicates the median distance between pre-selected and recorded geolocations across all 20 shehias

In our baseline mapping survey, almost all (99.7%) pre-selected housing structures were identified at the spot, in the communities, and located with a median distance of 5.4 m between the randomized centroids of the housing structures and the geolocation recorded by the enumerator. The computer-based randomization and offline navigation app-based geolocation of housing structures worked hence very well, even in remote villages in Pemba where "houses" may be small in size and located closely to each other and where internet connectivity is often limited or completely absent. The correct and exact identification of prospectively randomized housing structures is important for the accurate implementation of a community-based cross-sectional survey and also enables an easy follow-up of individuals or households for treatment or in longitudinal studies. Moreover, highly accurate small-scale spatial data can help to link household participant infection data with possible transmission sites of schistosomiasis, and hence to directly identify or model high-risk zones for infection and transmission that require special attention and targeted interventions. The location data can also be used to assess the distance to the next primary health care unit. A study conducted in rural Zambia showed that the preciseness of geographical cluster locations is crucial to link individuals to the correct health provider, such as primary health care units [34].

The dropout rate due to non-residential housing structures in our survey was 21.3%, absence of inhabitants was 9.0%, refused participation of households was 3.6%, and inability to provide a sufficiently large urine sample among all study participants was 15.5%. A very similar dropout rate of 24.1% due to uninhabited houses was observed in a household study based on spatial randomization from rural and urban regions of Cameroon [35]. The absence of inhabitants in our study was, however,

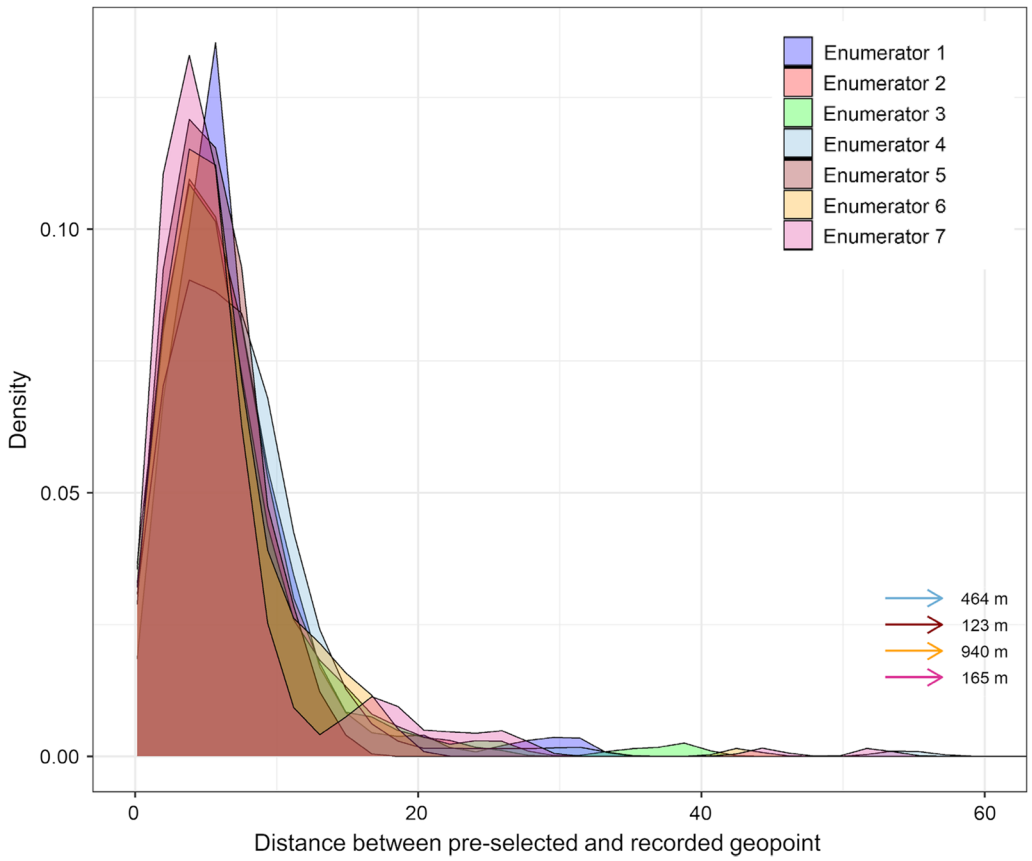


Fig. 4 Distance between pre-selected and recorded geolocations of housing structures in the mapping survey in Pemba, Tanzania, per enumerator

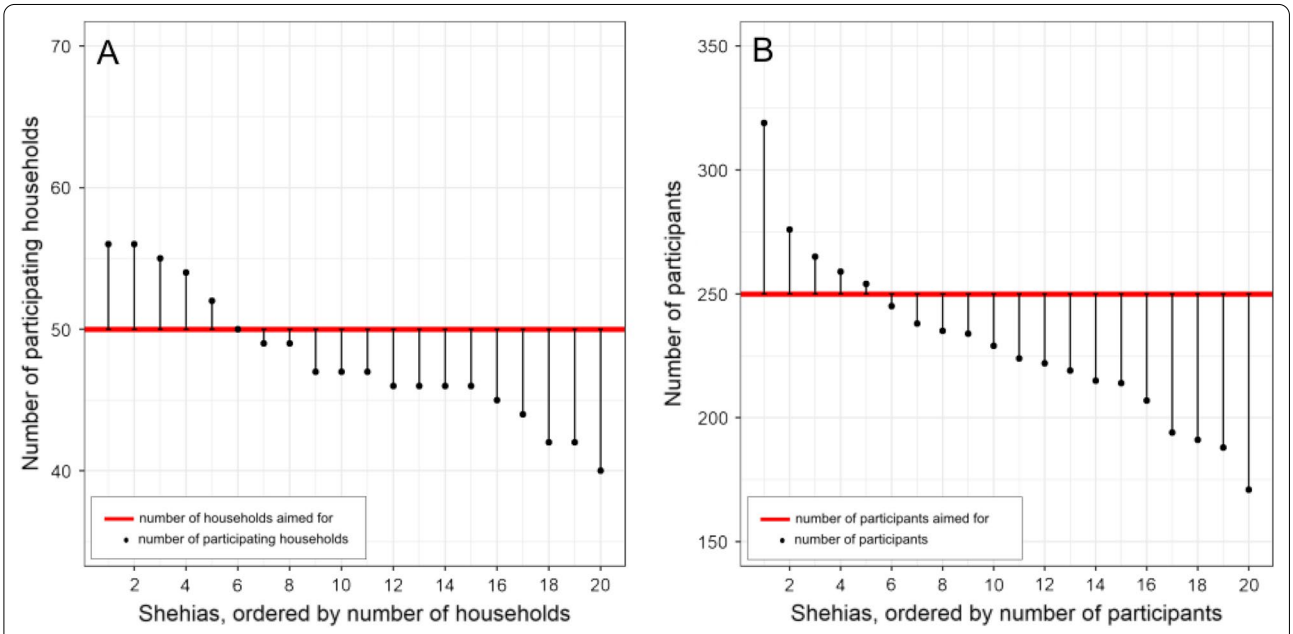


Fig. 5 Number of households (**A**) and individuals (**B**) participating in the mapping survey in Pemba, Tanzania, per shehia

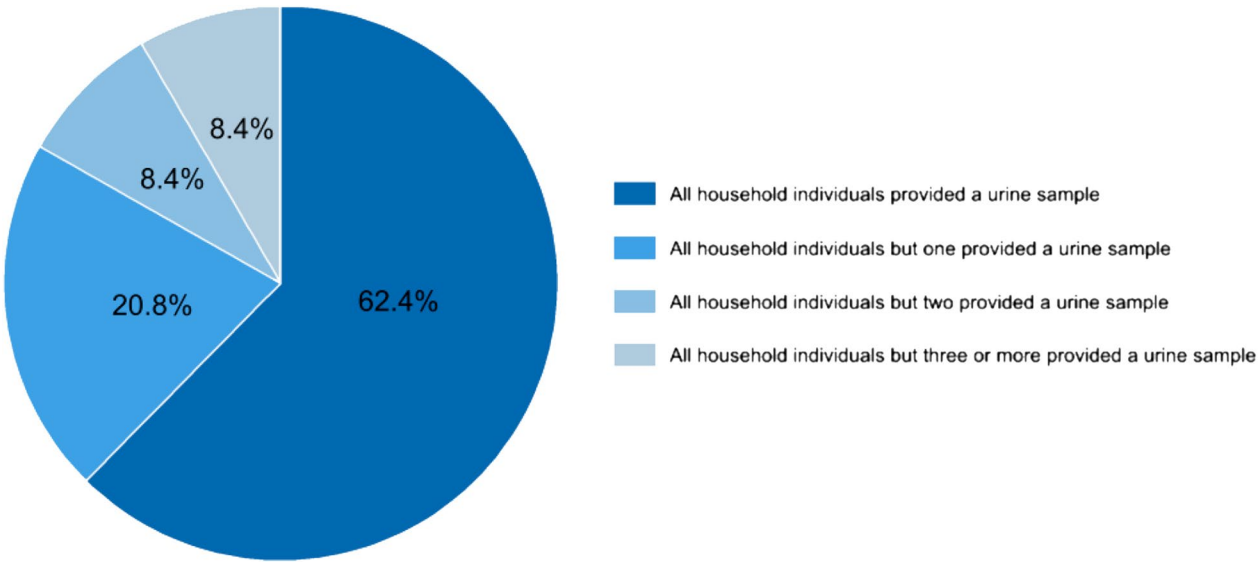


Fig. 6 Provision of urine samples. Percentage of households with study participants providing a urine sample of sufficient volume (≥ 10 ml) for schistosomiasis testing in the fine-scale mapping survey of the SchistoBreak study in Pemba, Tanzania

considerably lower than in a comparable GPS-based household survey conducted in Haiti where the absence of inhabitants was 16.4% [36]. This lower rate in our study might be explained by the fact that our enumerators visited the residential houses in a shehia for a second day if no individuals were at home the first day. The refusal dropout rate of 3.6% was, however, the same in our study as in the GPS-based household study conducted in Haiti [36]. A considerably higher dropout rate of 29.0% due to absence and/or inability to provide a urine and stool sample was observed in a household survey in Yemen, which was conducted to assess the local schistosomiasis prevalence [37]. A reason for the higher dropout in this study might be that not only urine but also stool samples were collected, which is more embarrassing to produce, collect and submit. In summary, our results show that our originally estimated overall dropout rate of 30% due to housing structures not being residential houses or refusal of household members to participate in the study (versus the actually observed dropout rate of 31.5%) was realistic and can serve as a guiding value for future survey design planning and sample size calculations, provided that 2 or 3 days are planned for participant recruitment and sample collection. In case households are only visited on a single day, higher dropout rates need to be considered in schistosomiasis mapping surveys, due to the potential absence of household members, or their inability or unwillingness to provide a urine or stool sample straight away. This being said, for *S. haematobium* diagnosis, urine samples are ideally collected in the late morning or early afternoon, when egg excretion is highest [38]. If urine samples can be produced and kept for the enumerator at any time of the day or night, this might impact on the diagnostic results and hamper accurate disease mapping.

With our practical introduction to and documentation of geolocation-based household identification and participant recruitment, we provide a rigorous basis for future survey planning. Particular strengths of our suggested approach are that it is low-cost, precise and easy to implement in the field as the final ODK questionnaire on the mobile device includes an automated link to the offline navigation system MAPS.ME. Due to the fully offline procedure it can also be used in remote areas without access to stable internet connections. Second, if data of housing structures are available on OSM, the fine-scale mapping approach is easy to replicate in other countries and for other purposes, particularly with the hands-on explanation provided in this article.

However, there are also important limitations. First, the approach relies on the availability of up-to-date (housing) data uploaded on OSM. In Pemba, new housing structures appear and disappear within a short time, or might be "under construction" for several years as has also been observed in other environments [36]. Without current data that include all new housing structures, individuals living in these housing structures will not be included in the randomization and missed for disease mapping. Without current data that mark or deleted demolished housing structures, the dropout rate will increase. To complement a higher dropout of housing structures if such invalid housing structures still appear in the data,

the sample size could be increased or replacement-housing structures could be included in the survey, as done in other studies [35, 39]. Second, our approach relies on the accessibility of the housing structures. For example, four housing structures could not be included in our survey due to inaccessibility, mainly as they were located in a military area. The data that provided the basis for the randomization of housing structures did not show the military area and therefore the area was not excluded prior to randomization. Hence, the success of our mapping approach is very much depending on the availability of up-to-date high-resolution data, clearly indicating zones with restricted access.

Conclusions

We show that the fine-scale mapping approach with the combination of ODK and an offline navigation application installed on tablet computers allows a very precise identification of pre-randomized housing structures. Dropouts due to non-residential houses, absence, non-participation and lack of urine need to be considered in sample size calculations. Our results can guide the planning and implementation of future household-based mapping or longitudinal surveys and thus support micro-targeting and follow-up of interventions for schistosomiasis control and elimination in remote areas.

Abbreviations

EKNZ: Ethikkommission Nordwest- und Zentralschweiz; MDA: Mass drug administration; NTD: Neglected tropical disease; ODK: Open Data Kit; OSM: Open Street Map; Swiss TPH: Swiss Tropical and Public Health Institute; WHO: World Health Organization; ZAHRI: Zanzibar Health Research Institute.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s40249-021-00928-y.

Additional file 1. Supplementary data that support the findings of this study from Nov 2020 until Feb 2021.

Additional file 2. Supplementary data dictionary that explains the data collected from Nov 2020 until Feb 2021.

Acknowledgements

We thank all household members who participated in our household survey from November 2020 until February 2021 and the field-enumerators in Pemba who conducted the household survey. Aurelio Di Pasquale is acknowledged for his repeated support with the ODK software and server at Swiss TPH. We are grateful for the information kindly provided by Mario Andrés Gronert Álvarez, which helped us to link our ODK questionnaires with MAPS.ME. Furthermore, we thank the Mapping Section, Department of Survey and Urban Planning, Zanzibar Commission for Lands for the shapefiles containing information about shehia borders and housing structures in the North of Pemba.

Authors' contributions

LT, SaMA, JH and SK conceptualized the SchistoBreak study and SaMA and SK initiated the SchistoBreak study. LT performed the data curation, formal analysis and data visualization for the research article. SK acquired the funding for the SchistoBreak study. MNA contributed significantly to the data collection and implementation of the household survey. LT, JH and SK conceptualized the statistical methods of the study. LT, ShMA, SaMA, FK and SK were responsible for the management of the research activity planning. LT conducted the randomization of housing structures and the connection of ODK with MAPS.ME for the household survey. LT, ShMA, SaMA and SK mentored the core team and MNA, ShMA, SaMA and FK advised for local conditions. LT and SK have drafted the first version of the research article. All authors have made substantial contributions to the study, and have reviewed and approved the submitted version of the manuscript.

Funding

Funding for the study has been obtained from the Swiss National Science Foundation (SNSF) via the PRIMA grant (PR00P3_179753) of Stefanie Knopp. This funding source had no role in the design of this study and will not have any role during its execution, analyses, interpretation of the data, or decision to submit results.

Availability of data and materials

Most relevant data are within the manuscript and its additional information files. Geopoints of the housing structures' centroids and geolocations recorded by the field enumerators cannot be shared to preserve spatial confidentiality.

Declarations

Ethics approval and consent to participate

The study protocol has been waived by the ethics committee of Switzerland (Ethikkommission Nordwest- und Zentralschweiz; EKNZ) on October 23, 2019 (Req-2019-00951) and has been approved by the ethics committee of Zanzibar (Zanzibar Health Research Institute; ZAHRI) on December 13, 2019 (ZAHREC/02/November/2019/16). All study participants submitted an informed consent form signed by a parent or legal guardian in case of participating children and adolescents, or signed by the participant in case of participating adults. The study was prospectively registered at ISRCTN (ISCRCTN91431493).

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author details

[1]Swiss Tropical and Public Health Institute, Kreuzstrasse 2, 4123 Allschwil, Switzerland. [2]University of Basel, Petersplatz 1, 4001 Basel, Switzerland. [3]Public Health Laboratory-Ivo de Carneri, Wawi, P.O. Box 122, Chake-Chake, Pemba, United Republic of Tanzania. [4]Neglected Diseases Program, Zanzibar Ministry of Health, Social Welfare, Elderly, Gender and Children, P.O. Box 236, Unguja, United Republic of Tanzania.

Received: 2 November 2021 Accepted: 17 December 2021
Published online: 15 January 2022

References

1. WHO. Ending the neglect to attain the Sustainable Development Goals—a road map for neglected tropical diseases 2021–2030. Geneva: World Health Organization; 2020.
2. WHO. Schistosomiasis key facts 2020. https://www.who.int/news-room/fact-sheets/detail/schistosomiasis. Accessed 15 Dec 2021.
3. GBD 2017 DALYs and HALE Collaborators. Global, regional, and national disability-adjusted life-years (DALYs) for 359 diseases and injuries and healthy life expectancy (HALE) for 195 countries and territories, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet. 2018;392(10159):1859–922.
4. Steinmann P, Keiser J, Bos R, Tanner M, Utzinger J. Schistosomiasis and water resources development: systematic review, meta-analysis, and estimates of people at risk. Lancet Infect Dis. 2006;6(7):411–25.

5. Colley DG, Bustinduy AL, Secor WE, King CH. Human schistosomiasis. Lancet. 2014;383(9936):2253–64.
6. Deol AK, Fleming FM, Calvo-Urbano B, Walker M, Bucumi V, Gnandou I, et al. Schistosomiasis—assessing progress towards the 2020 and 2025 global goals. N Engl J Med. 2019;381(26):2519–28.
7. Webster JP, Molyneux DH, Hotez PJ, Fenwick A. The contribution of mass drug administration to global health: past, present and future. Philos Trans R Soc Lond B Biol Sci. 2014;369(1645):20130434.
8. Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies. PLoS Negl Trop Dis. 2021;15(2):e0009127.
9. Rollinson D, Knopp S, Levitz S, Stothard JR, Tchuem Tchuenté LA, Garba A, et al. Time to set the agenda for schistosomiasis elimination. Acta Trop. 2013;128(2):423–40.
10. King CH, Kittur N, Binder S, Campbell CH, N'Goran EK, Meite A, et al. Impact of different mass drug administration strategies for gaining and sustaining control of *Schistosoma mansoni* and *Schistosoma haematobium* infection in Africa. Am J Trop Med Hyg. 2020;103(1_Suppl):14–23.
11. Zhou Y, Chen Y, Jiang Q. History of human schistosomiasis (bilharziasis) in China: from discovery to elimination. Acta Parasitol. 2021;66(3):760–9.
12. Santos IGA, Bezerra LP, Cirilo TM, Silva LO, Machado JPV, Lima PD, et al. New epidemiological profile of schistosomiasis from an area of low prevalence in Brazil. Rev Soc Bras Med Trop. 2020;53:e20200335.
13. Souza MDR, de Jesus DMS, Santos AHC, Lima S, Dos Santos A, Tavares DDS, et al. Risk clusters of *Schistosoma mansoni* infection in an endemic state of Brazil: space-time modelling and association with socio-economic and environmental factors. Trans R Soc Trop Med Hyg. 2021. https://doi.org/10.1093/trstmh/trab090.
14. Fornace KM, Fronterrè C, Fleming FM, Simpson H, Zoure H, Rebollo M, et al. Evaluating survey designs for targeting preventive chemotherapy against *Schistosoma haematobium* and *Schistosoma mansoni* across sub-Saharan Africa: a geostatistical analysis and modelling study. Parasit Vectors. 2020;13(1):555.
15. Shen Y, Wiegand RE, Olsen A, King CH, Kittur N, Binder S, et al. Five-year impact of different multi-year mass drug administration strategies on childhood *Schistosoma mansoni*-associated morbidity: a combined analysis from the Schistosomiasis Consortium for Operational Research and Evaluation cohort studies in the Lake Victoria regions of Kenya and Tanzania. Am J Trop Med Hyg. 2019;101(6):1336–44.
16. Phillips AE, Tohon Z, Dhanani NA, Sofo B, Gnandou I, Sidikou B, et al. Evaluating the impact of biannual school-based and community-wide treatment on urogenital schistosomiasis in Niger. Parasit Vectors. 2020;13(1):557.
17. Camara Y, Sanneh B, Joof E, Sanyang AM, Sambou SM, Sey AP, et al. Mapping survey of schistosomiasis and soil-transmitted helminthiases towards mass drug administration in The Gambia. PLoS Negl Trop Dis. 2021;15(7):e0009462-e.
18. Mawa PA, Kincaid-Smith J, Tukahebwa EM, Webster JP, Wilson S. Schistosomiasis morbidity hotspots: roles of the human host, the parasite and their interface in the development of severe morbidity. Front Immunol. 2021;12:635869.
19. Wiegand RE, Mwinzi PNM, Montgomery SP, Chan YL, Andiego K, Omedo M, et al. A persistent hotspot of *Schistosoma mansoni* infection in a five-year randomized trial of praziquantel preventative chemotherapy strategies. J Infect Dis. 2017;216(11):1425–33.
20. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-year intervention study on elimination of urogenital schistosomiasis in Zanzibar: parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019;13(5):e0007268.
21. Menghistu HT, Mersha TT, Shumuye NA, Woldie BM, Redda YT, Hadush B, et al. Neglected tropical zoonotic diseases in Tigray region, northern Ethiopia: spatial distribution and trend analysis of rabies, tuberculosis, schistosomiasis, and visceral leishmaniasis in humans. Zoonoses Public Health. 2021;68(7):823–33.
22. Hatherell H-A, Simpson H, Baggaley RF, Hollingsworth TD, Pullan RL. Sustainable surveillance of neglected tropical diseases for the post-elimination era. Clin Infect Dis. 2021;72(Suppl 3):S210–6.
23. Tchuem Tchuenté LA, Stothard JR, Rollinson D, Reinhard-Rupp J. Precision mapping: an innovative tool and way forward to shrink the map, better target interventions, and accelerate toward the elimination of schistosomiasis. PLoS Negl Trop Dis. 2018;12(8):e0006563.
24. Soares Magalhães RJ, Salamat MS, Leonardo L, Gray DJ, Carabin H, Halton K, et al. Geographical distribution of human *Schistosoma japonicum* infection in The Philippines: tools to support disease control and further elimination. Int J Parasitol. 2014;44(13):977–84.
25. Monnier N, Barth-Jaeggi T, Knopp S, Steinmann P. Core components, concepts and strategies for parasitic and vector-borne disease elimination with a focus on schistosomiasis: a landscape analysis. PLoS Negl Trop Dis. 2020;14(10):e0008837.
26. Fornace KM, Surendra H, Abidin TR, Reyes R, Macalinao MLM, Stresman G, et al. Use of mobile technology-based participatory mapping approaches to geolocate health facility attendees for disease surveillance in low resource settings. Int J Health Geogr. 2018;17(1):21.
27. Wagenaar BH, Augusto O, Ásbjörnsdóttir K, Akullian A, Manaca N, Chale F, et al. Developing a representative community health survey sampling frame using open-source remote satellite imagery in Mozambique. Int J Health Geogr. 2018;17(1):37.
28. Trippler L, Hattendorf J, Ali SM, Ame SM, Juma S, Kabole F, et al. Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study. BMC Infect Dis. 2021;21(1):1024.
29. Office of Chief Government Statistician Zanzibar. Zanzibar statistical abstract 2019. 2020.
30. Office of Chief Government Statistician Zanzibar. Zanzibar in figures 2019. 2020.
31. Office of Chief Government Statistician Zanzibar. 2012 Population and housing census: population distribution by administrative areas. 2013.
32. Hatz C, Savioli L, Mayombana C, Dhunputh J, Kisumku UM, Tanner M. Measurement of schistosomiasis-related morbidity at community level in areas of different endemicity. Bull World Health Organ. 1990;68(6):777–87.
33. Savioli L, Mott KE. Urinary schistosomiasis on Pemba Island: low-cost diagnosis for control in a primary health care setting. Parasitol Today. 1989;5(10):333–7.
34. Carter ED, Munos MK. Impact of imprecise household location on effective coverage estimates generated through linking household and health provider data by geographic proximity: a simulation study. Int J Health Geogr. 2021;20(1):38.
35. Alcoba G, Ochoa C, Babo Martins S, Ruiz de Castañeda R, Bolon I, Wanda F, et al. Novel transdisciplinary methodology for cross-sectional analysis of snakebite epidemiology at national scale. PLoS Negl Trop Dis. 2021;15(2):e0009023.
36. Wampler PJ, Rediske RR, Molla AR. Using ArcMap, Google Earth, and Global Positioning Systems to select and locate random households in rural Haiti. Int J Health Geogr. 2013;12:3.
37. Sady H, Al-Mekhlafi HM, Mahdy MAK, Lim YAL, Mahmud R, Surin J. Prevalence and associated factors of schistosomiasis among children in Yemen: implications for an effective control programme. PLoS Negl Trop Dis. 2013;7(8):e2377.
38. Scott JA, Stimmel CM. The regularity of egg output of *Schistosoma haematobium*. Tex Rep Biol Med. 1956;14(4):440–58.
39. Kondo MC, Bream KDW, Barg FK, Branas CC. A random spatial sampling method in a rural developing nation. BMC Public Health. 2014;14:338.

3.5. Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: baseline cross-sectional survey findings before the onset of a 3-year intervention study

Lydia Trippler[1,2], Said Mohammed Ali[3], Shaali Makame Ame[3], Jan Hattendorf[1,2], Khamis Rashid Suleiman[3], Mohammed Nassor Ali[3], Saleh Juma[4], Fatma Kabole[4], Stefanie Knopp[1,2]

1 Swiss Tropical and Public Health Institute, Allschwil, Switzerland

2 University of Basel, Basel, Switzerland

3 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

4 Neglected Diseases Programme, Zanzibar Ministry of Health, Social Welfare, Elderly, Gender and Children, Lumumba, Unguja, United Republic of Tanzania

This article was published in:

Parasites & Vectors (2022), Aug 16; 15(1):292

doi: 10.1186/s13071-022-05404-6

Parasites & Vectors

RESEARCH Open Access

Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: baseline cross-sectional survey findings before the onset of a 3-year intervention study

Lydia Trippler[1,2], Said Mohammed Ali[3], Shaali Makame Ame[3], Jan Hattendorf[1,2], Khamis Rashid Suleiman[3], Mohammed Nassor Ali[3], Saleh Juma[4], Fatma Kabole[5] and Stefanie Knopp[1,2*]

Abstract

Background: Schistosomiasis elimination has gained renewed priority in the WHO guidance documents published in 2020 and 2022. The SchistoBreak project, implemented in Pemba, Tanzania between 2020 and 2024, aims to assess new tools and strategies for shifting from elimination as a public health problem towards interruption of transmission. Here we report our baseline findings and discuss implications for future interventions.

Methods: In 2020, human water contact sites (HWCSs) in the study area were geolocated and snail surveys were conducted. A parasitological and questionnaire cross-sectional baseline survey was implemented in 20 communities and their 16 primary schools between November 2020 and February 2021. Urine samples were collected at the school and household levels from individuals aged ≥ 4 years. *Schistosoma haematobium* infection was detected by urine filtration microscopy. Snail, parasitological and questionnaire-derived data were analyzed descriptively, spatially and with generalized estimated equation models.

Results: The intermediate host snail *Bulinus globosus* was detected in 19.8% (33/167) of HWCSs. The overall *S. haematobium* prevalence was 1.2% (26/2196) in school-aged children and 0.8% (31/3893) in community members, with 0.2% (4/2196) and 0.1% (3/3893) heavy-intensity infections, respectively. Children who studied < 1 km away from HWCSs with *B. globosus* had significantly higher odds for a *S. haematobium* infection than those attending a school located > 2 km away (odds ratio [OR]: 5.0; 95% confidence interval [CI]: 2.3–11.1). Individuals living in a house located < 1 km away from HWCSs with *B. globosus* had higher odds than those residing in > 2 km distance (OR: 18.0; 95% CI: 2.9–111.0). Self-reported praziquantel treatment coverage was 83.2% (2015/2423) in schoolchildren in the mass drug administration (MDA) conducted in August 2020. Coverage among adult community members was 59.9% (574/958), but only 34.8% (333/958) took praziquantel correctly.

*Correspondence: s.knopp@swisstph.ch

[1] Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, Allschwil, Switzerland
Full list of author information is available at the end of the article

Conclusions: While the *S. haematobium* prevalence is very low in Pemba, there are many HWCSs with *B. globosus* situated close to schools or houses that pose a considerable risk of recrudescence. To maintain and accelerate the progress towards interruption of transmission, targeted and cost-effective interventions that are accepted by the community are needed; for example, snail control plus focal MDA, or test-and-treat in schools and households near infested waterbodies.

Keywords: *Bulinus globosus*, Distance, Elimination, Fine-scale mapping, Freshwater bodies, Interruption of transmission, Mass drug administration, *Schistosoma haematobium*, Surveillance response, Test-and-treat

Introduction

Schistosomiasis is a highly debilitating disease endemic in 78 countries worldwide [1]. Its highest burden is concentrated in sub-Saharan Africa, where 91% (42/46) of the countries are affected [2]. Recent model-based estimates indicate, however, that the schistosomiasis prevalence in sub-Saharan Africa has decreased considerably over the past few decades, likely due to the scale-up of preventive chemotherapy programs [3].

With the "Roadmap for neglected tropical diseases 2021–2030" and the "Guideline on control and elimination of human schistosomiasis" published by the WHO in 2020 and 2022, respectively, the global elimination of schistosomiasis as a public health problem by 2030 and the interruption of transmission in selected countries by 2030 gained renewed priority and important guidance [1, 4].

Pemba Island, part of the Zanzibar archipelago in the United Republic of Tanzania, has achieved elimination of urogenital schistosomiasis as a public health problem since 2017 [5–7]. Continued actions are required to sustain and even advance these gains made towards interruption of transmission [4]. The SchistoBreak project, implemented in 20 communities in the north of Pemba Island in 2020 and ongoing until 2024, aims to assess new tools and strategies for exactly this purpose [8]. Largely in accordance with the new WHO recommendations 2, 4 and 5 [4], three strategies are implemented in the smallest administrative areas (shehias) of the study area dependent on prevalence: (i) shehias with a *Schistosoma haematobium* prevalence $\geq 3.0\%$ in the annual school-based survey and/or $\geq 2.0\%$ in the annual community-based survey will receive regular preventive chemotherapy plus snail control and behavior change measures to further reduce transmission and infection prevalence; ii) shehias with a *S. haematobium* prevalence $< 3.0\%$ in the annual school-based survey and $< 2.0\%$ in the annual community-based survey will receive targeted test-treat-track interventions of at-risk populations plus focal snail control to prevent recrudescence; and (iii) all shehias implement passive surveillance and treatment of cases in their health facilities [8].

The results of the SchistoBreak study will generate important evidence to support and potentially reshape existing guidelines and provide new insights into interrupting *S. haematobium* transmission in Pemba and other low-prevalence settings in the future. Here, we report the baseline findings from 2020/2021 and discuss implications for present and future risk assessments and intervention approaches.

Methods

Study setting

The SchistoBreak study is being conducted on Pemba Island, which is part of the Zanzibar archipelago in the Indian Ocean, located around 30 km east of the Tanzanian mainland. Pemba is divided into four main districts, which are further split at the sub-district level into 129 small administrative areas, called shehias [9]. Our study area covers 20 shehias, which in this paper are referred to as "implementation units" (IUs), across the two districts of Micheweni and Wete in the north of Pemba. Urogenital schistosomiasis, caused by *S. haematobium*, used to be highly endemic in Pemba in the 1980s [10, 11]. No other *Schistosoma* species infecting humans are known to be endemic on the Zanzibar islands [12]. Interventions to control morbidity of urogenital schistosomiasis in Pemba started with test-and-treat activities in the 1980s [13] and later expanded to regular island-wide mass drug administration (MDA) campaigns in the early 2000s [14]. More intense elimination efforts were carried out as part of a cluster-randomized trial on islands of the archipelago, Unguja and Pemba, between 2011/2012 and 2017 [5, 7]. The most recent results published from Pemba indicate a low overall *S. haematobium* prevalence of 3.4% among schoolchildren aged 9–12 years and 0.4% among adults aged 20–55 years in 2020 [6]. A considerable temporal and spatial heterogeneity exists, however, with many low-prevalence areas and a few remaining hotspots [5–7].

Characteristics of IUs included in the survey

To gain a comprehensive picture of the educational and environmental characteristics of the study area, a survey was conducted in the 20 study IUs between February and July 2020, with a 3-month interruption due to COVID-19

that resulted in a lockdown by the Tanzanian government from March to June 2020. Details on the IU characteristics survey have been published elsewhere [8]. In brief, initial interviews were held with the leader (sheha) of each IU who provided information on the respective IU. Data were collected on the population size and number of housing structures, the number and type of schools and the number and locations of natural open freshwater bodies and public clean water sources. Subsequently, each nursery, primary and secondary school and all Madrassas (Islamic schools) in the IUs were visited and data were collected on each school's geolocation and the number of children enrolled in 2020. Finally, all known human water contact sites (HWCSs) were located in the IUs, their geolocation recorded and data collected on the type and characteristics of the water bodies and the abundance of freshwater snail species—specifically the intermediate-host snail *Bulinus globosus*. Individuals who were present at the HWCSs at the time of the visit were invited to share their opinion on their community's needs for better protection against *S. haematobium* infections.

Additionally, in September 2021, all health facilities used by individuals living in the IUs were visited, their geolocation recorded and data collected about the type of the health facility (primary health care unit [PHCU] vs hospital, and public vs private). All data were captured with Open Data Kit (ODK) software (www.opendatakit.org) installed on Samsung Galaxy Tab A tablets (Samsung Electronics, Seoul, South Korea).

Cross-sectional parasitological surveys

To assess the prevalence of *S. haematobium* infections in Pemba at baseline, a cross-sectional parasitological survey was conducted in the 20 study IUs from November 2020 until March 2021. Based on the survey results, the IUs were stratified into hotspot IUs or low-prevalence IUs to assign respective measures for the intervention period following the baseline survey. The survey included a school-based and a community-based component to obtain an accurate picture of *S. haematobium* infections in: (i) the school-aged population, which is considered to be at the highest risk of infection [15], and (ii) the whole population, including all individuals aged ≥ 4 years, in the study areas.

School-based survey

The school-based cross-sectional survey was conducted in the largest public primary school of each IU [8]. All children aged ≥ 4 years enrolled in the school were eligible to participate in the study. Each school was visited on 2 consecutive days. On day 1, one class (A, B, C, D…) in each of seven grades (nursery school, and grade 1 to grade 6) was selected based on a computer-generated randomized list. In each selected class, the students were asked to line up, with girls and boys lining up separately, and each third child in the lines was selected until a total of 25 children per class, half girls, half boys, were included in the study. Demographic information, such as names, age and sex, was collected from all selected students. The selected children were also asked about any travel in the previous 6 months and about their participation in the school-based MDA with praziquantel conducted in August 2020. Each selected child received a consent and information form to be signed by their parent or legal guardian and to be returned the following day. Children aged ≥ 12 years were additionally asked to provide their own written assent for participation. On day 2, between 9 AM and 2 PM, a urine collection cup was provided to all children who had returned a signed consent form. Each cup was labeled with a unique identifier code. The children were advised to fill their cup with their own fresh urine sample and subsequently submit it to the field enumerators.

Community-based survey

The community-based cross-sectional survey was conducted in each of the 20 study IUs [8]. In preparation for the survey, 70 housing structures per IU were randomly selected from available shape file data provided by the Zanzibar Commission for Lands to the Zanzibar Neglected Diseases Program [16]. Field enumerators identified the geolocation of each selected housing structure with ODK, combined with the offline navigation application "MAPS.ME" (www.maps.me). Details of the randomization process of housing structures and the GPS-based fine-scale mapping approach are described elsewhere [16]. Each IU was visited for 3 consecutive days. On day 1, the enumerators visited all selected housing structures in the IU, explained the aims of the study in Kiswahili to all household members present at the time and invited them, as well as those family members who were absent at the time but who were returning to the residence later in the day, to participate. Once written informed consent was obtained, an interview with one adult household member who was present was conducted. The questionnaire covered sociodemographic details (age, name, sex) of all household members, as well as information on participation of the interviewed person in the last community-based MDA, their opinion on MDA as a measure against schistosomiasis and their travels in the previous 6 months. All household members aged ≥ 4 years were eligible to submit a urine sample for *S. haematobium* testing. For this purpose, an information and consent form was provided to each eligible household member to be signed by themselves if they were ≥ 18 years of age or else by their parent or

legal guardian. Children aged ≥ 12 years were additionally asked to provide their own written assent for participation. Moreover, each eligible household member was given a plastic cup for urine collection that was labeled with a unique identifier code and a sticker with a specific drawing (for example of a boat, cat, or cow). The stickers were linked to the same stickers on a paper list, where the names of the household members were indicated in addition to the stickers to allow a clear association of drawing and name and hence to avoid confusion of the urine cups between the household members. All consenting participants were asked to fill the container with one sample of their own urine until the next morning. On day 2, the enumerators revisited the households from day 1 for urine container collection and for collection of signed consent and assent forms. All residential houses where nobody was present on day 1 were revisited. On day 3, all signed consent and assent forms and urine cups that had not been submitted on day 1 or 2 were collected.

Laboratory examinations

All urine samples were transferred to the Public Health Laboratory—Ivo de Carneri (PHL-IdC) in Wawi, ChakeChake for analysis on the same day. Each sample was examined for microhematuria using reagent strips (Hemastix; Siemens Healthcare Diagnostics AG; Zurich, Switzerland). Additionally, each sample was filtered through a 13-mm fabric filter (Sefar Ltd., Bury, UK) placed in a Swinnex plastic filter holder (Millipore-Merck KgaA, Darmstadt, Germany) using a 10-ml plastic syringe and examined for the presence and number of *S. haematobium* eggs under a light microscope.

Data management

All laboratory results for both the school- and community-based surveys were captured on paper and double entered onto electronic spreadsheets (Excel 2010; Microsoft Corp., Redmond, WA, USA). Double entered data were compared and cleaned with R version 4.0.3 (www.rproject.org) and Stata/IC 16.1 (StataCorp LLC, College Station, TX, USA). Discrepant results were verified against data on the corresponding original paper forms and re-entered correctly. All registration and questionnaire data captured in ODK were sent to the ODK central server hosted at the Swiss Tropical and Public Health Institute in Basel, Switzerland. For statistical analyses, coded laboratory results were merged with the registration and/or questionnaire information using R and Stata/IC. Participant names were kept in a separate file and only relinked with the coded results to inform all participants about their infection status with *S. haematobium*.

Statistical methods

Statistical analyses were performed using R version 4.0.3. The median number of public water taps reported by the sheha was determined per 1000 IU inhabitants and rounded to whole number.

The geolocations of nursery, primary and secondary schools and Madrassas identified in the IU characteristics survey, the number of children enrolled in the schools and all identified HWCSs in the study area together with the number of *B. globosus* found were mapped with ArcMap version 10.6.1 (ESRI, Redlands, CA, USA).

All eligible individuals who provided a urine sample were included in the parasitological analysis. A participant was considered to be *S. haematobium*-positive if ≥ 1 egg was detected by urine filtration. *Schistosoma haematobium* infection was classified into two intensity categories: (i) light intensity (1–49 *S. haematobium* eggs/10 ml), and (2) and heavy intensity (≥ 50 *S. haematobium* eggs/10 ml) [17].

A participant was considered to be microhematuria-positive if blood in urine was detected by reagent strips, with the microhematuria graded as trace, +, ++ or +++ based on the color chart provided by the manufacturer of the strips. *Schistosoma haematobium* prevalence was calculated overall as well as at both the individual school and community levels. An IU was stratified as a hotspot IU if the *S. haematobium* prevalence in the school was ≥ 3.0% and/or ≥ 2.0% in the corresponding community. An IU was considered as a low-prevalence IU if the *S. haematobium* prevalence in the school was < 3.0% and < 2.0% in the corresponding community.

To predict the spatial *S. haematobium* prevalence, the study area was divided into 5350 equally sized grid points, and the prevalence per 1 km radius around the center of each grid point was determined by dividing the number of *S. haematobium*-positive individuals by the total number of community members surveyed in the 1-km radius around the center of the grid point. All HWCSs, both those with and without detected *B. globosus*, were plotted on a map indicating the predicted *S. haematobium* prevalence per 1-km radius around the center of each grid point.

To estimate the odds ratios (ORs) for a *S. haematobium* infection, generalized estimating equation (GEE) models with exchangeable correlation structure were run. Separate models were applied for the school-based survey and the community-based survey. Independent variables included in the models for both surveys were sex, age groups and distance from the school or residential house, respectively, to the next HWCS with *B. globosus*. In addition, in the model for the school-based survey, travel and reported treatment in the past 6 months were included as

independent variables. In the model for the community-based survey, distance from the residential house to the next health facility was included as an additional independent variable. In the models, 95% confidence intervals (CI), and GEE with robust standard errors to account for clustering were used. Stratified by the school-based survey and community-based survey, either the schools or the communities were included in the models as clusters.

To determine the treatment coverage of the MDA conducted in August 2020 and participants' intake of praziquantel, assessments were made of how many participants reported to have received and taken treatment in the school-based or community-based MDA, respectively. Moreover, the survey also assessed how many participants had received praziquantel at least once in their lifetime and if they considered MDA with praziquantel as a good or "not-good" intervention to fight schistosomiasis. The narrative responses were translated into English and grouped into thematic categories.

Results

Implementation Unit characteristics survey

The she has of the 20 study IUs reported population sizes ranging from 1030 to 7000 inhabitants, with a median of 4393. The number of housing structures reported ranged from 513 to 1400, with a median of 782. The number of public water taps per 1000 IU inhabitants reported by the she has was between zero and 74, with a median of six.

A total of 367 schools were visited in the 20 IUs, of which 23 (6.3%) were primary schools, with 21 (91.3%) being public schools and two(8.7%) being private schools; 16 (4.4%) were secondary schools, with 14 (88.0%) being public schools and two (12.5%) being private schools; 89 (24.3%) were nursery schools, with 86 (96.6%) being public schools and three (3.4%) being private schools; and 239 (64.9%) were Madrassas. The location and number of enrolled students at each school is indicated in Fig. 1A.

A total of 167 HWCSs were identified in the 20 study IUs, including 112 (67.1%) located at water bodies characterized as rivers or streams and 55 (32.9%) located at lakes or ponds. *Bulinus globosus* was found in 33 (19.8%) of the 167 HWCSs. The location of HWCSs and the number of *B. globosus* collected is indicated in Fig. 1B.

At 47 (28.1%) of the 167 identified HWCSs, people were present and willing to share their opinion on the needs of the community for better protection against *S. haematobium* infections. The most highlighted need was to "*apply medicine to the water*" for reducing the amount of snails or simply to "*kill the snails*", mentioned by people at 29 (61.7%) of the 47 HWCSs.

In the 20 study IUs, there were 21 health facilities, including 14 (66.7%) and five (23.8%) public and private PHCUs, respectively, and two (9.5%) public hospitals (Fig. 1B).

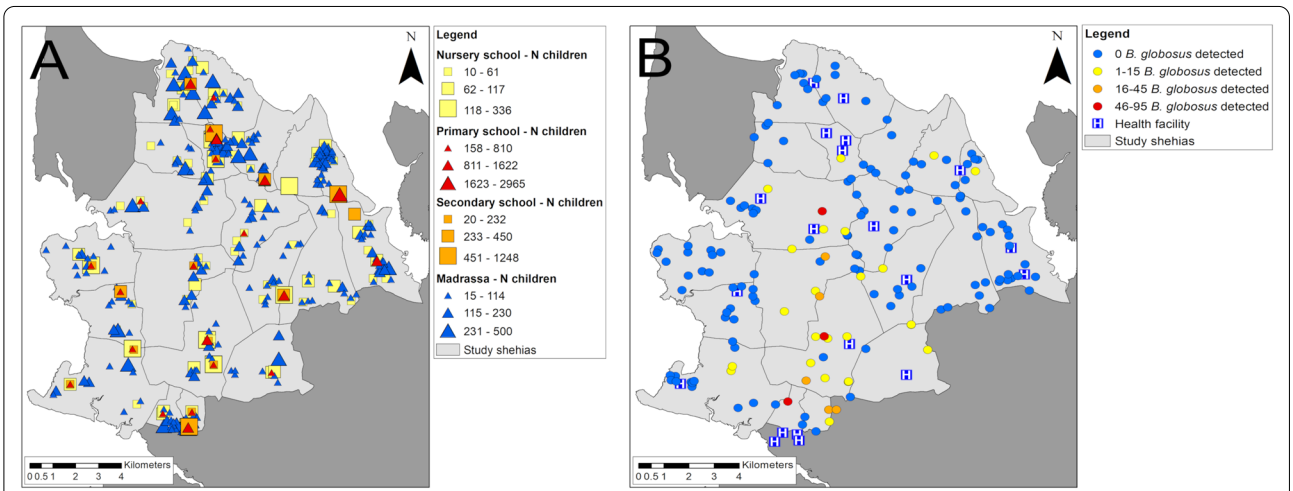


Fig. 1 Schools, human water contact sites (HWCSs) and health facilities in the study area in Pemba, Tanzania. **A** The type and geolocations of schools with the number of children enrolled, **B** the geolocations of HWCSs with the number of *Bulinus globosus* detected and the geolocations of health facilities within the SchistoBreak study area in the north of Pemba, Tanzania. The image base map (United Republic of Tanzania—subnational administrative boundaries) was downloaded from the United Nations (UN) Office for the Coordination of Humanitarian Affairs (OCHA) services (https://data.humdata.org/dataset/tanzania-administrative-boundaries-level-1-to-3-regions-districts-and-wards-with-2012-population). The data source of the image base map is Tanzania National Bureau of Statistics/UN OCHA ROSA. The data of the image base map are published under the following license: Creative Commons Attribution for Intergovernmental Organisations (CC BY-IGO; (https://creativecommons.org/licenses/by/3.0/igo/legal code)). Written permission was obtained to use and adapt the data from OCHA. Additional shape files for the map (shehia boundaries) were provided by the Zanzibar Commission for Lands to the Zanzibar Neglected Diseases Program

Cross-sectional parasitological survey

Study participation

In the school-based survey, a total of 2465 children from 16 schools were randomly selected and invited to participate (Fig. 2). Among these, 227 (9.4%) were absent on the day of urine collection, and 42 (1.7%) refused to participate or did not provide a signed consent form. In total, urine samples were collected and analyzed from 2196 children, of whom 1167 (53.1%) were female and 1029 (46.9%) were male (median age: 10 years, range 4–17 years).

In the community-based survey, 1400 housing structures were randomly selected (Fig. 2). Of these, 441 (31.5%) were not sampled because the structures were located in protected military areas with prohibited access, were not residential houses, had no household members at home during the time of survey or all household members refused to participate. Ultimately, 959 residential houses with a total of 4599 household members were included in the study. Among the 4599 household members, 705 (15.3%) were excluded from the analyses because of ineligibility due to their age (< 4 years old) or missing urine samples. Hence, 3893 individuals were included in the final community-based analysis, of whom 2067 (53.0%) were female and 1826 (47.0%) were male (median age: 18 years, (range: 4–87 years)) .

Schistosoma haematobium infection and microhematuria in schools

Among the 2196 children included in the analyses of the school-based survey, 26 (1.2%) children tested positive for *S. haematobium* by urine filtration, with heavy-intensity infections in four (0.2%) students (median egg count: 15, range 2–94). The median age of *S. haematobium*-positive children was 11 (range: 4–15) years. Among the 1167 female and 1029 male participants, 12 (1.0%) and 14 (1.4%), respectively, tested positive for *S. haematobium*. In three of the 16 surveyed schools, the *S. haematobium* prevalence was higher than the pre-set 3.0% threshold for classifying IUs as hotspots (Fig. 3A). Among the 2196 children participating in the school-based survey, 78 (3.6%) were microhematuria-positive with a median age of 11 (range: 4–15) years. Among the 1167 girls and 1029 boys, 43 (3.7%) and 35 (3.4%) tested positive for microhematuria, respectively.

Schistosoma haematobium infection and microhematuria in communities

Among the 3893 individuals participating in the community-based survey, 31 (0.8%) individuals tested positive for *S. haematobium*, with a heavy-intensity infection occurring in three (0.1%) individuals (median egg count of six, range 1–168). The median age of *S. haematobium*-positive participants was 10 years (range 6–52). Among the 2067 female participants, 15 (0.7%) tested *S. haematobium*-positive and among the 1826 male participants, 16 (0.9%) tested *S. haematobium*-positive. In three of the 20 IU communities, the *S. haematobium* prevalence was higher than the pre-set 2.0% threshold for classifying IUs as hotspots (Fig. 3B). Among the 3893

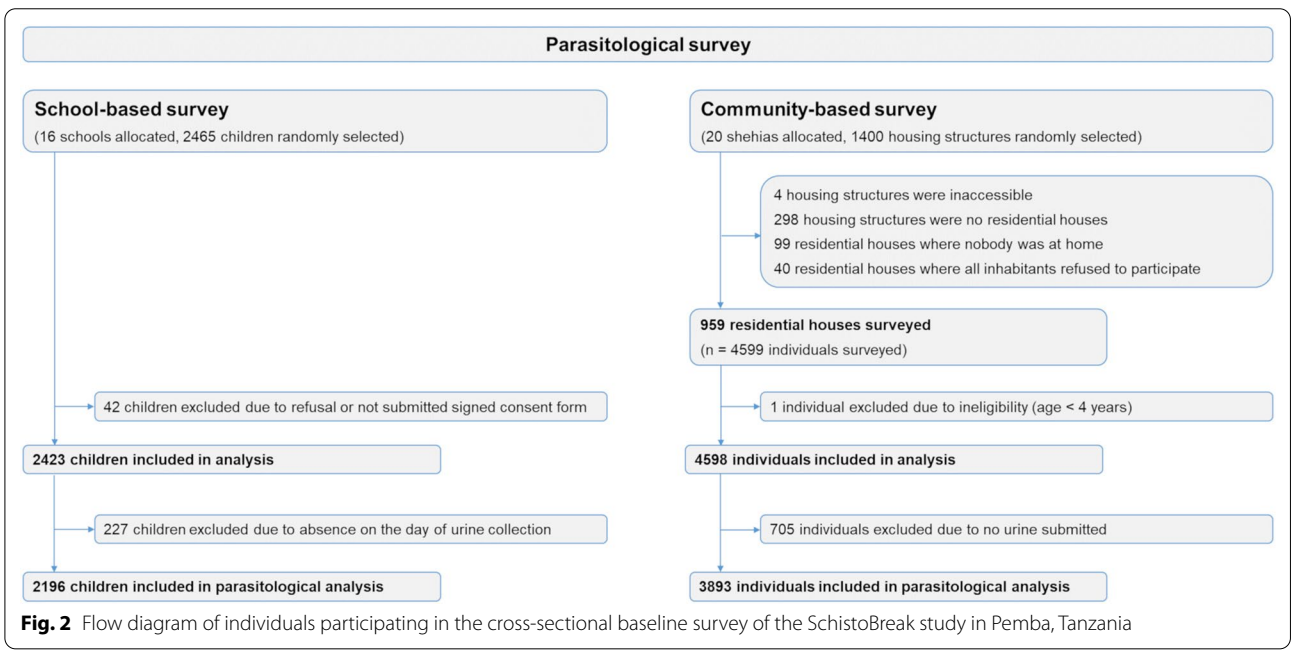


Fig. 2 Flow diagram of individuals participating in the cross-sectional baseline survey of the SchistoBreak study in Pemba, Tanzania

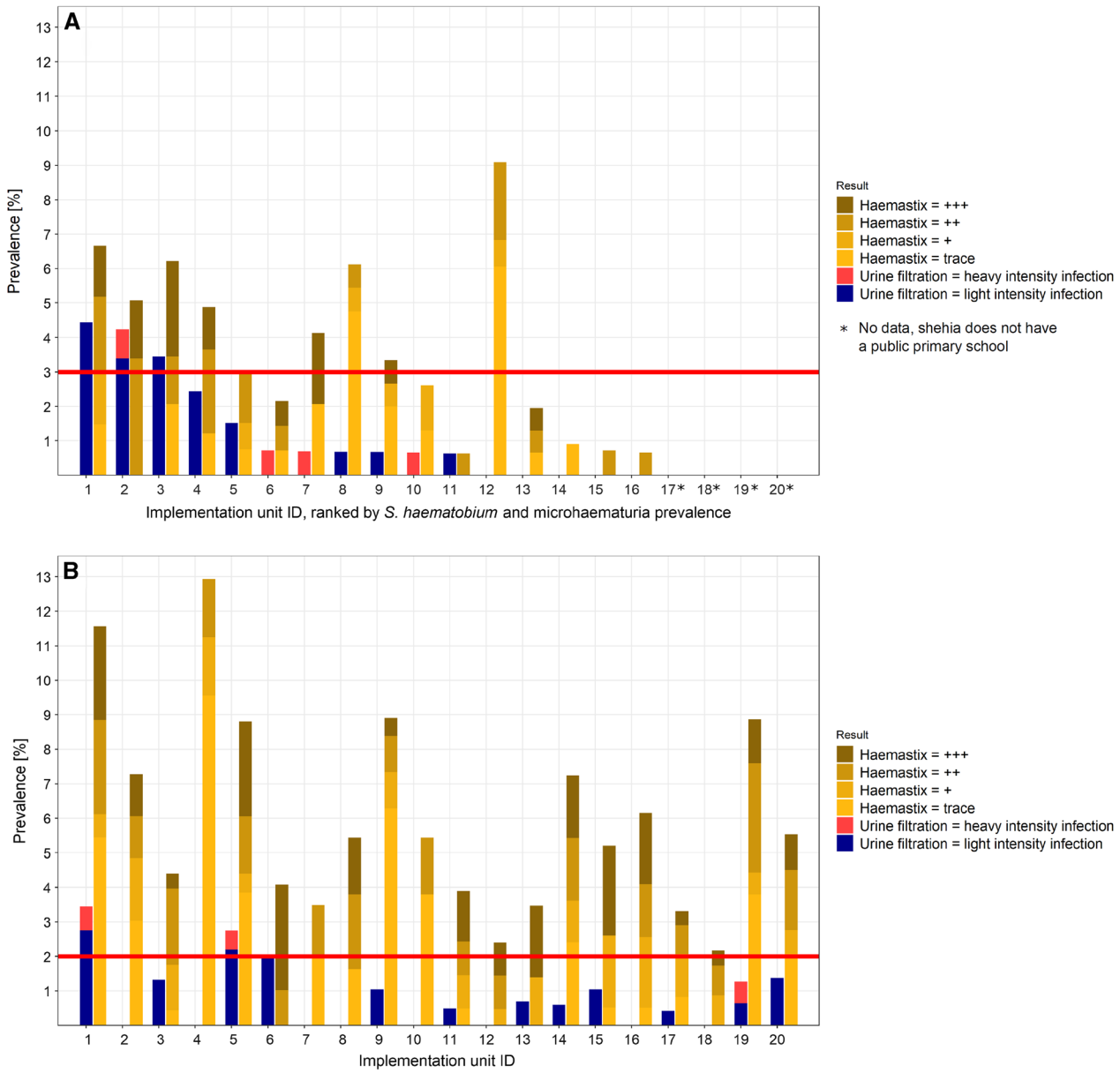


Fig. 3 Baseline *Schistosoma haematobium* and microhematuria prevalence in the implementation units (IUs) of the SchistoBreak study in Pemba, Tanzania. **A**, **B** *Schistosoma haematobium* and microhematuria prevalence stratified by schools (**A**) and communities (**B**) in the 20 IUs of the study area in the north of Pemba, Tanzania. The ID numbers of the schools and communities correspond to the same IU. Asterisks denote those IUs (ID numbers 17, 18, 19, 20) with no main primary school to be included in the survey. Microhematuria was graded as trace, +, ++ or +++ based on the color chart provided by the manufacturer of the haemastix test strips. The red line indicates the threshold for the stratification of the IUs into hotspot or low-prevalence IUs based on urine filtration results

individuals participating in the community-based survey, 225 (5.8%) were microhematuria-positive with a median age of 20 years (rage 4–85). Among the 2067 female and 1826 male participants, 154 (7.5%) and 71 (3.9%) were microhematuria-positive, respectively.

Stratification in hotspot and low-prevalence implementation units

As indicated in Fig. 3A, B and Fig. 4A, three schools and three communities, in a total of five IUs, crossed the $\geq 3.0\%$ and/or $\geq 2.0\%$ prevalence threshold for

S. haematobium infections, respectively. Our baseline survey therefore defined a total of five hotspot IUs and 15 low-prevalence IUs, respectively, for the intervention period following the survey in 2021 (Fig. 4B).

Geo-spatial distribution of S. haematobium infection and B. globosus in the study area

A geo-spatial analysis of the *S. haematobium* prevalence in the community-based survey per 1-km radius around the center of equally distributed grid points revealed that 64.8% of the geographical study area had a predicted *S. haematobium* prevalence of 0.0% (Fig. 5). In another 24.4% of the geographical study area, the predicted *S. haematobium* prevalence was > 0.0% and < 10.0%. In 0.5% of the geographical study area, the predicted *S. haematobium* prevalence was ≥ 10.0%. No *S. haematobium* data were available for the remaining 10.3% of the study area as it was uninhabited land.

The highest *S. haematobium* prevalence observed within a 1-km radius around one of the grid points was 18.2%. The median distance between a grid point plus a 1-km radius with a *S. haematobium* prevalence ≥ 1.0% and a HWCSs with *B. globosus* was 877 m. The median distance between a grid point plus a 1-km radius with a *S. haematobium* prevalence < 1.0% and HWCSs with *B. globosus* was 1616 m.

Risk factors for S. haematobium infection in schools

In the school-based survey, a total of 2194 participants were included in the GEE model analyses. No statistically significant association was revealed between *Schistosoma haematobium* infection and sex (Fig. 6A). Children aged 13–17 years had a significantly higher chance (OR: 2.9; CI: 1.1–7.6; prevalence: 2.5% *versus* 1.1%) of acquiring a *S. haematobium* infection than children aged 4–8 years. Children aged 9–12 years did not have a higher chance of being infected with *S. haematobium* than children aged 4–8 years. Moreover, children enrolled in a school located < 1 km away from the closest HWCS where *B. globosus* was detected had significantly higher odds (OR: 5.0; 95% CI: 2.3–11.1; prevalence: 2.4% vs 0.5%) of being infected with *S. haematobium* than children enrolled in a school that was located > 2 km away from the closest HWCS with *B. globosus*. The OR for children enrolled in a school 1–2 km away from the closest HWCS where *B. globosus* was detected was 1.0 (95% CI: 0.3–3.2; prevalence: 0.5%) compared with children enrolled in a school > 2 km away from the closest HWCS with *B. globosus*. No statistically significant association was determined for travel in the last 6 months. Children who had not received praziquantel during the previous 6 months had 2.4-fold higher odds (95% CI:

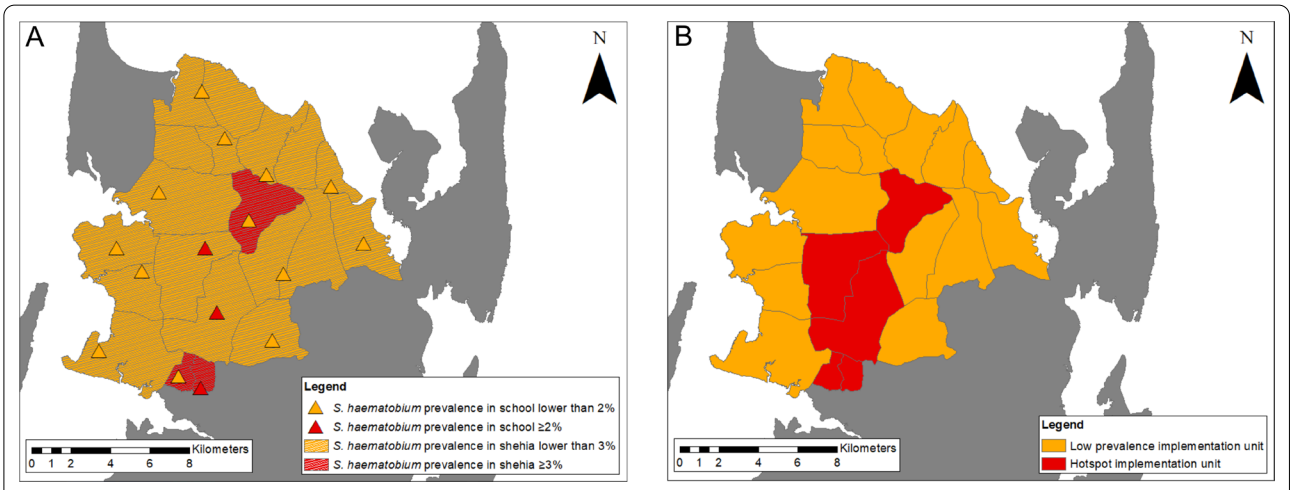


Fig. 4 *Schistosoma haematobium* prevalence in the study sites and their classification into hotspot and low-prevalence implementation units (IUs). **A** *Schistosoma haematobium* prevalence in study schools and communities as identified during the cross-sectional parasitological survey from November 2020 to March 2021 below or above pre-defined thresholds. **B** Classification of the IUs into hotspot and low-prevalence IUs based on data shown in **A**. The image base map (United Republic of Tanzania—subnational administrative boundaries) was downloaded from the UN Office for the Coordination of Humanitarian Affairs (OCHA) services (https://data.humdata.org/dataset/tanzania-administrative-boundaries-level-1-to-3-regions-districts-and-wards-with-2012-population). The data source is: Tanzania National Bureau of Statistics/UN OCHA ROSA. The data are published under the following license: Creative Commons Attribution for Intergovernmental Organisations (CC BY-IGO; https://creativecommons.org/licenses/by/3.0/igo/legal code). Written permission was received to use and adapt the data from OCHA. Additional shape files for the map (shehia boundaries) were provided by the Zanzibar Commission for Lands to the Zanzibar Neglected Diseases Program

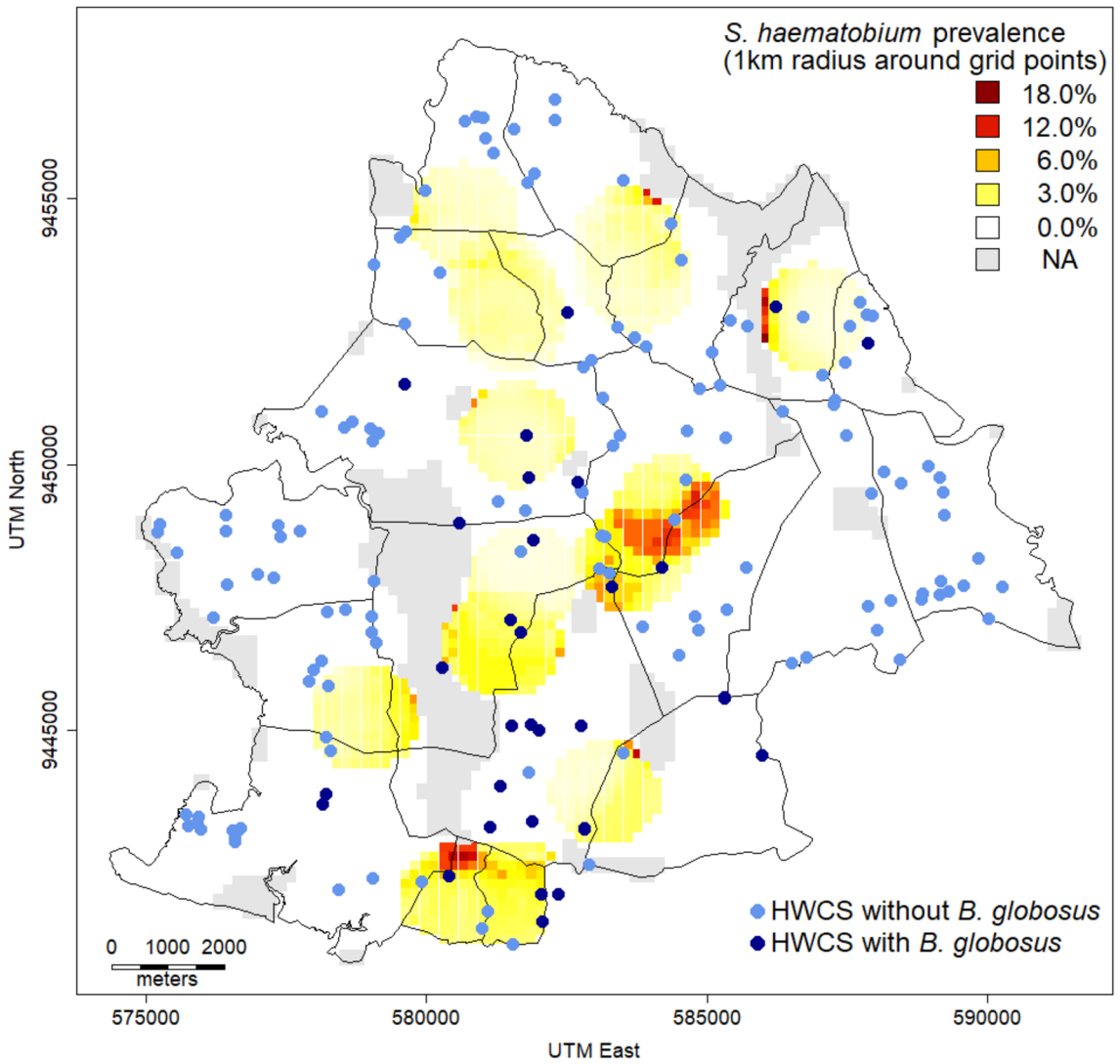


Fig. 5 Geo-spatial distribution of *Schistosoma. haematobium* prevalence in the community-based survey and Human water contact sites (HWCSs) with and without *Bulinus globosus*. The figure shows the *S. haematobium* prevalence per 1-km radius around the center of equally sized grid points observed during the community-based survey and the spatial distribution of HWCSs with and without *B. globosus* detected during the implementation unit (IU) characteristics survey of the SchistoBreak study in the north of Pemba, Tanzania. The shape files for the map (shehia boundaries) were provided by the Zanzibar Commission of Lands to the Zanzibar Neglected Diseases Program

(See figure on next page.)

Fig. 6 Multivariable analysis of risk factors for *Schistosoma haematobium* infection in participants of the SchistoBreak baseline survey.**A**,**B** The odds ratios (ORs) for a Schistosoma *haematobium* infection adjusted for various risk factors observed within the cross-sectional school-based (**A**) and community-based (**B**) surveys conducted from November 2020 to March 2021 in the north of Pemba, Tanzania.

a1) indicates the distance from the school to the nearest human watercontact site(HWCS) where *Bulinus globosus* was detected during the implementation unit (IU) characteristics survey.

a2) indicates the distance from the residential house of the individual to the nearest human water contact sites (HWCS) HWCS where *B. globosus* was detected during the IU characteristics survey.

b) indicates the distance from the residential house of the individual to the health facility. Abbreviations:

Prev., prevalence

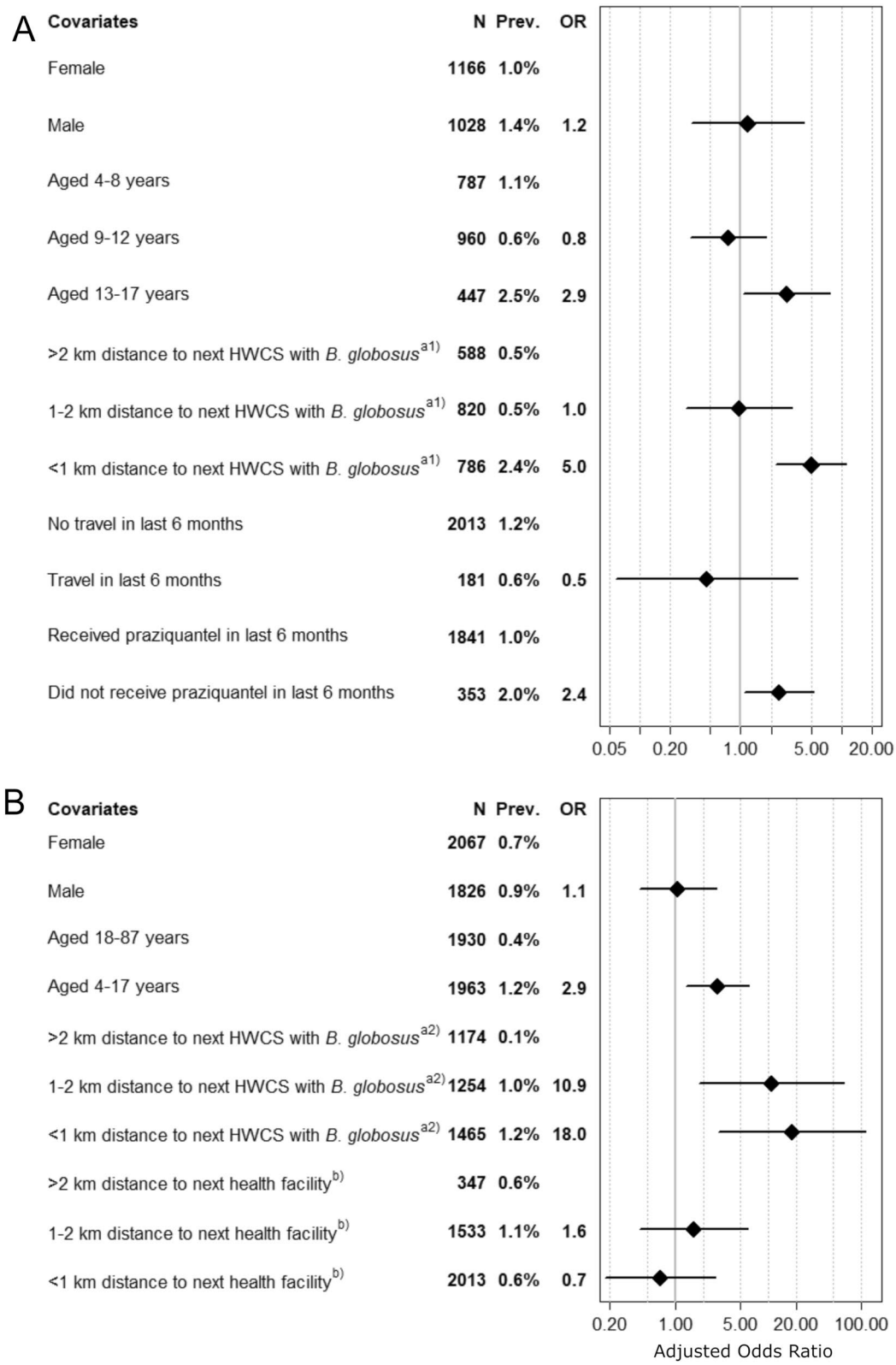


Fig. 6 (See legend on previous page.)

1.1–5.3; prevalence: 2.0% vs 1.0%) of being infected with *S. haematobium* than children who had received treatment in the previous 6 months.

Risk factors for S. haematobium infection in communities

In the community-based survey, a total of 3893 individuals were included in the GEE model analyses. No statistically significant association was determined between *S. haematobium* infection and sex (Fig. 6B). Children (4–17 years) had significantly higher odds of a *S. haematobium* infection (OR: 2.9; 95% CI: 1.3–6.2; prevalence: 1.2% vs 0.4%) than adults (aged ≥ 18 years). Individuals residing 1–2 km from a HWCS with *B. globosus* had a significantly higher chance of a *S. haematobium* infection (OR: 10.9; 95% CI: 1.8–64.4; prevalence: 1.0% vs 0.1%) than individuals living > 2 km away from the next HWCS with *B. globosus*. The odds for a *S. haematobium* infection were highest in individuals who resided < 1 km away from a HWCS with *B. globosus* (OR: 18.0; 95% CI: 2.9–111.0; prevalence: 1.2% vs 0.1%) compared with individuals living > 2 km away from a HWCS with *B. globosus*. No statistically significant association was observed between a *S. haematobium* infection and the distance of residential houses to the next health facility.

MDA coverage and perception of MDA as a good or not-good intervention

In the school-based survey, 2015/2423 (83.2%) children reported that they were treated with praziquantel during the last school-based MDA in August 2020.

In the community-based survey, 574/958 (59.9%) individuals reported that they had received praziquantel during the last community-based MDA in August 2020 (Fig. 7). Among those 574 individuals who reported to have received the drug, 510 (88.9%) claimed to have swallowed the received tablets, with 333 (65.3%) individuals reporting that they took all of the tablets at the same time, 104 (20.4%) reporting that they took the tablets over multiple days, 62 (12.2%) reporting that they split the tablet intake to the morning and evening of the same day and 11 (2.2%) not answering the question. Among the 64 individuals who did not swallow the tablets, the two most common reasons reported for not swallowing praziquantel were being pregnant (13/64; 20.3%) and feeling healthy (5/64; 7.8%). Among all 958 adult participants interviewed, 333 (34.8%) received praziquantel and took all tablets at the same time, in line with WHO recommendations [4].

Moreover, 786/958 (82.0%) interviewed participants reported that they had received praziquantel at some time in their life. Among those who had ever received

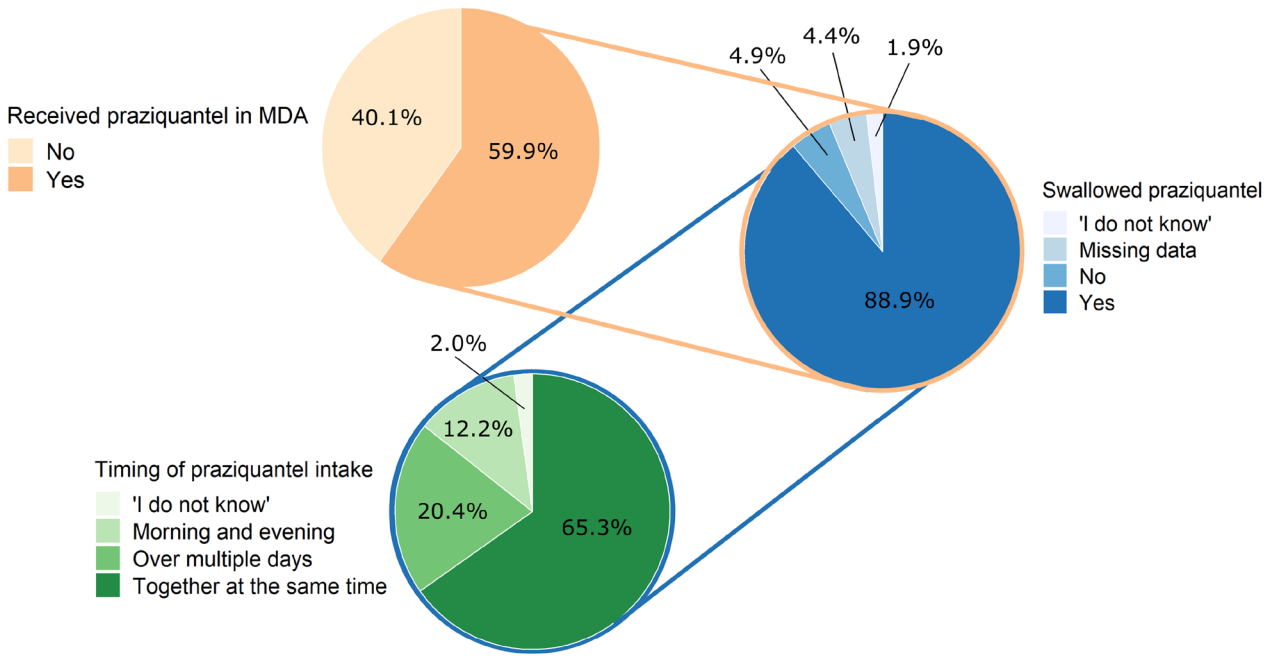


Fig. 7 Coverage and praziquantel intake among adults during the community-based mass drug administration (MDA) in August 2020

praziquantel in their life, 548 (69.7%) perceived MDA as a good intervention, 87 (11.1%) perceived MDA as not a good intervention, 143 (18.2%) were indecisive and eight (1.0%) did not answer. The most common thematic reasons for considering MDA a good intervention were "*It is good for the health*" (327/501; 65.3%) and "*One saves time/money/stress of going to the hospital to get the drugs as the drugs are delivered at home*" (74/501; 14.8%). The major reasons why MDA was not perceived as good were "*One is not tested for schistosomiasis before receiving the treatment*" (56/78; 71.8%) and "*The drug distributors are no medical personnel*" (6/78; 7.7%).

Discussion

Schistosomiasis elimination has been given a high priority in the WHO guidance documents published in 2020 and 2022 [1, 4]. Schistosomiasis as a public health problem has been eliminated on Pemba Island since 2017 [5, 6]. As is part of the Zanzibar archipelago, United Republic of Tanzania, Pemba is included on the WHO list of countries that have been targeted for interruption of schistosomiasis transmission by 2030. The SchistoBreak project implemented in the north of Pemba between 2020 and 2024 aims to assess new tools and strategies for moving from elimination as a public health problem towards interruption of transmission [8, 16].

The results of the baseline survey confirmed a low overall *S. haematobium* prevalence and percentage of heavy infection intensities, respectively, in both schools (1.2% and 0.2%) and communities (0.8% and 0.1%) of the SchistoBreak study area. The community-based survey revealed that in most (89.2%) of the geographical study area, the prevalence was <10.0% and that, according to the new WHO guidelines, the area can be considered to be a low-prevalence setting [4].

There is a large number of freshwater bodies in the study area, and many of these are infested with the intermediate host snail *B. globosus*. As such, these freshwater bodies are a risk for transmission and a challenge for elimination [18, 19]. The GEE model confirmed that children attending a school or community members living in a house within a 1-km radius of a HWCS with *B. globosus* had a 5- or 18-fold higher chance of being infected with *S. haematobium*, respectively. Our results are in line with those of other studies conducted in Zanzibar and Nigeria that also observed a positive relationship between *S. haematobium* infection and decreasing distance to HWCSs with *B. globosus* or freshwater bodies in general [3, 20–22].

These findings can have considerable implications for cost-effective schistosomiasis risk assessment and micro-targeting interventions in close-to-elimination settings: to identify potential high-transmission of *S. haematobium* sites, rigorous and repeated snail surveys at HWCSs should be conducted. Then, if *B. globosus* is detected, focal snail control measures could be applied coupled with focal MDA or test-and-treat targeting individuals living or attending schools within a 2-km radius around these HWCSs. Snail control measures involving niclosamide is recommended by WHO as a cost-effective intervention tool that has a large impact on *S. haematobium* infections [4, 23–25]. Many individuals (61.7%) interviewed at the HWCSs during our IU characteristics survey proposed applying "medicine," i.e. niclosamide, to the HWCSs to target the intermediate host snails, which indicates a high level of community acceptance for snail control in the study area. Focal snail control has been implemented successfully in other settings, such as Egypt and Mali [26, 27]. In similar low-prevalence settings for malaria, focal MDA plus vector control have been applied to reduce transmission [28, 29] and reactive case detection, and focal MDA against malaria were shown to have a high community acceptance in Namibia [28, 29].

There is a long history of large-scale MDA in Pemba and in other countries as preventive chemotherapy is recommended by WHO as an intervention to control morbidity of schistosomiasis and to advance towards the elimination of this disease as a public health problem in communities with a *Schistosoma* prevalence ≥ 10% [4, 10, 30]. The continuation of regular preventive chemotherapy is also recommended among existing control programs as they move towards interruption of transmission, such as in Pemba [4]. While the self-reported coverage and compliance with directly observed praziquantel intake in the school-based MDA in 2020 was 83.8% among schoolchildren in our study in Pemba, treatment coverage among adults in the community-based door-to-door approach was only 59.9%. Ultimately, only 34.8% of the targeted adult population received and swallowed all of the tablets at the same time—and hence as a single oral dose—in line with WHO recommendations [4], which is worryingly low and might point to treatment fatigue. The main reasons for not swallowing any of the received praziquantel tablets were pregnancy and feeling healthy at the time. All adult participants who ever participated in an MDA in the past were asked about their opinion about MDA. Among those who considered MDA not as a good intervention, their qualitative response was "*MDA is not a good intervention because one is not tested for schistosomiasis before receiving the treatment.*" Hence, offering praziquantel in health facilities, including mother and child health care units, by medical personnel [31] and using a test-and-treat strategy instead of preventive chemotherapy without prior diagnosis might increase

compliance among adults. However, to date, no sensitive and specific test for *S. haematobium* detection at the point-of-care exists, presenting a considerable challenge for low-prevalence settings where most people are only slightly infected [32–36]. The development of a highly sensitive and specific test that is affordable and simple and can also be used in health facilities in low-prevalence areas is an urgent need when moving towards interruption of transmission and post-elimination surveillance [37–40].

In the SchistoBreak study, we will explore some of the intervention approaches recommended by WHO in their new guidelines and/or those suggested above. We are confident that our study will reveal important insights into and provide evidence for their feasibility and contribution to the goal of interrupting transmission. While the baseline survey already has produced valuable results, there are a number of limitations worth highlighting. First, the snail surveys during the IU characteristics survey were conducted only once per HWCS. Hence, there may be HWCSs where *B. globosus* was not detected during the time of visit but which are a transient habitat for the snails, possibly distorting our results. Second, *S. haematobium* prevalence was determined by a single urine filtration per person. The low sensitivity of the test to detect light-intensity infections could result in an underestimation of the true prevalence [36]. Third, in the school-based survey, the GEE model was based on the distance of the children's school to HWCSs with *B. globosus* and not the children's homes, which might have provided more accurate results than the distance from the school.

Conclusions

We found that the overall prevalence of *S. haematobium* in the north of Pemba is low, but the many HWCSs with *B. globosus* situated close to schools or houses represent a high risk for rebounding transmission and recrudescence of infection. Furthermore, while treatment coverage among schoolchildren was high, the correct intake of praziquantel during MDA was low among adults. To maintain the gains made and accelerate towards interruption of *S. haematobium* transmission in Pemba and elsewhere, there is a need for new processes and integrated intervention strategies that are cost-effective and accepted in the community, such as intensified snail surveys, focal snail control, focal MDA and/or targeted test-and-treat approaches.

Abbreviations

CI: Confidence interval; GEE: Generalized estimating equation; HWCS: Human water contact site; IU: Implementation unit; MDA: Mass drug administration; ODK: Open data kit; OR: Odds ratio; PHCU: Primary health care unit.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13071-022-05404-6.

Additional file 1: Additional data that support the findings of schools within the IU characteristics survey from February 2020 until July 2020 (CSV).

Additional file 2: Additional data dictionary that explains the data of schools collected during the IU characteristics survey from February 2020 until July 2020 (TXT).

Additional file 3: Additional data that support the findings of water bodies within the IU characteristics survey from February 2020 until July 2020 (CSV).

Additional file 4: Additional data dictionary that explains the data of water bodies collected during the IU characteristics survey from February 2020 until July 2020 (TXT).

Additional file 5: Additional data that support the findings of implementation units within the IU characteristics survey from February 2020 until July 2020 (CSV).

Additional file 6: Additional data dictionary that explains the data of implementation units collected during the IU characteristics survey from February 2020 until July 2020 (TXT).

Additional file 7: Additional data that support the findings of health facilities within the IU characteristics survey in September 2021 (CSV).

Additional file 8: Additional data that explains the data of health facilities collected during the IU characteristics survey in September 2021 (TXT).

Additional file 9: Additional data that support the findings of the baseline survey from November 2020 until February 2021 (CSV).

Additional file 10: Additional data dictionary that explains the data collected during the baseline survey from November 2020 until February 2021 (TXT).

Acknowledgements

We thank all shehas and teachers for welcoming us into their communities and for agreeing to collaborate with us during the SchistoBreak study period, for sharing information about their shehias and schools and for their support during the cross-sectional parasitological surveys. Moreover, we are happy that a collaboration between SchistoBreak and the local health facilities has started in 2020 and thank all employees of the health facilities for their cooperation. We feel special gratitude to every child and community member who participated in the baseline survey of SchistoBreak. Furthermore, we thank two independent reviewers and Rory Doolan for their comments and edits that helped to improve the content and English language of the manuscript.

Author contributions

LT, SaMA, JH and SK conceptualized the SchistoBreak study. SaMA and SK initiated the SchistoBreak study. LT, MNA and SK performed the data curation. LT and JH conducted the formal analysis and data visualization for the research article. SK acquired the funding for the SchistoBreak study. KRS and MNA contributed significantly to the data collection and implementation of the IU characteristics survey and the baseline survey. LT, JH and SK conceptualized the statistical methods of the study. LT, SaMA, ShMA, FK and SK were responsible for the management of the research activity planning. LT, SaMA, ShMA and SK mentored the core team and KRS, MNA, SaMA, ShMA, SJ and FK advised for local conditions. LT and SK have drafted the first version of the research article. All authors have made substantial contributions to the study. All authors read and approved the final manuscript.

Funding

Funding for the study has been obtained from the Swiss National Science Foundation (SNSF) via the PRIMA grant (PR00P3_179753/1) of Stefanie Knopp. This funding source had no role in the design of this study and will not have any role during its execution, analyses, interpretation of the data or decision to submit results.

Availability of data and materials
All data generated or analysed during this study are included in this published article and its Additional information files (Additional file 1, 2, 3, 4, 5, 6, 7, 8, 9, 10).

Declarations

Ethics approval and consent to participate
The protocol of the study was waived by the ethics committee of Switzerland (Ethikkommission Nordwest- und Zentralschweiz; EKNZ) on 23 October 2019 (Req-2019–00951) and approved by the ethics committee of Zanzibar (Zanzibar Health Research Institute; ZAHRI) on 13 December 2019 (ZAHREC/03/PR/December/2019/12). The protocol renewal and its amendment were approved by ZAHRI on 2 December 2020 (ZAHREC/01/RN/December/2020/10) and 23 February (ZAHREC/04/AMEND/FEB/2021/01), respectively. The study was prospectively registered at ISRCTN (ISCRCTN91431493). All individuals participating in the cross-sectional survey between November 2020 and March 2021 were informed in detail about the objectives and procedures of the survey and provided written informed consent for their participation. In case of participating children aged ≤ 18 years, written consent was obtained from their parents or legal guardians. Children aged 12–17 years additionally signed an assent form themselves.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

Author details
[1]Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, Allschwil, Switzerland. [2]University of Basel, Basel, Switzerland. [3]Public Health Laboratory—Ivo de Carneri, Wawi Pemba, United Republic of Tanzania. [4]Neglected Diseases Program, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania. [5]Neglected Tropical Diseases Program Ministry of Health , Lumumba, Unguja, United Republic of Tanzania.

Received: 9 May 2022 Accepted: 21 July 2022
Published online: 16 August 2022

References

1. WHO. Ending the neglect to attain the sustainable development goals—a road map for neglected tropical diseases 2021–2030. 2021. https://www.who.int/publications/i/item/9789240010352. Accessed on 02.08.2022
2. WHO. Schistosomiasis progress report 2001–2011, strategic plan 2012–2020. 2013. https://apps.who.int/iris/handle/10665/78074. Accessed on 02.08.2022
3. Kokaliaris C, Garba A, Matuska M, Bronzan RN, Colley DG, Dorkenoo AM, et al. Effect of preventive chemotherapy with praziquantel on schistosomiasis among school-aged children in sub-Saharan Africa: a spatiotemporal modelling study. Lancet Infect Dis. 2022;22:136–49. https://doi.org/10.1016/S1473-3099(21)00090-6.
4. WHO. WHO guideline on control and elimination of human schistosomiasis. 2022. https://www.who.int/publications/i/item/9789240041608. Accessed.
5. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-year intervention study on elimination of urogenital schistosomiasis in Zanzibar: parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019;13:e0007268. https://doi.org/10.1371/journal.pntd.0007268.
6. Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies. PLoS Negl Trop Dis. 2021;15:e0009127. https://doi.org/10.1371/journal.pntd.0009127.
7. Knopp S, Person B, Ame SM, Ali SM, Hattendorf J, Juma S, et al. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: a cluster-randomised trial. Lancet Glob Health. 2019;7:e1118–29. https://doi.org/10.1016/s2214-109x(19)30189-5.
8. Trippler L, Hattendorf J, Ali SM, Ame SM, Juma S, Kabole F, et al. Novel tools and strategies for breaking schistosomiasis transmission: Study protocol for an intervention study. BMC Infect Dis. 2021;21:1024. https://doi.org/10.1186/s12879-021-06620-8.
9. Office of Chief Government Statistician Zanzibar. Zanzibar in figures 2019. 2020. http://www.ocgs.go.tz/php/ReportOCGS/ZIF,%202019%20%20%20FINAL%20(14..8.2020%20for%20printing).pdf. Accessed on 02.08.2022
10. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba Island: programme organization and management. Trop Med Parasitol. 1989;40:189–94.
11. Savioli L, Mott KE. Urinary *schistosomiasis* on Pemba Island: low-cost diagnosis for control in a primary health care setting. Parasitol Today. 1989;5:333–7. https://doi.org/10.1016/0169-4758(89)90129-4.
12. Stothard JR, Mgeni AF, Khamis S, Seto E, Ramsan M, Rollinson D. Urinary schistosomiasis in schoolchildren on Zanzibar Island (Unguja), Tanzania: a parasitological survey supplemented with questionnaires. Trans R Soc Trop Med Hyg. 2002;96:507–14. https://doi.org/10.1016/S0035-9203(02)90421-9.
13. Savioli L, Dixon H, Kisumku UM, Mott KE. Control of morbidity due to *Schistosoma haematobium* on Pemba island; selective population chemotherapy of schoolchildren with haematuria to identify high-risk localities. Trans R Soc Trop Med Hyg. 1989;83:805–10. https://doi.org/10.1016/0035-9203(89)90336-2.
14. Guidi A, Andolina C, Makame Ame S, Albonico M, Cioli D, Juma HH. Praziquantel efficacy and long-term appraisal of schistosomiasis control in Pemba Island. Trop Med Int Health. 2010;15:614–8. https://doi.org/10.1111/j.1365-3156.2010.02488.x.
15. Hotez PJ, Kamath A. Neglected tropical diseases in sub-saharan Africa: review of their prevalence, distribution, and disease burden. PLoS Negl Trop Dis. 2009;3:e412. https://doi.org/10.1371/journal.pntd.0000412.
16. Trippler L, Ali MN, Ame SM, Ali SM, Kabole F, Hattendorf J, et al. GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation. Infect Dis Poverty. 2022;11:8. https://doi.org/10.1186/s40249-021-00928-y.
17. Montresor A, Crompton D, Hall A, Bundy D, Savioli L, et al. Guidelines for the evaluation of soil-transmitted helminthiasis and schistosomiasis at community level. 1998. https://apps.who.int/iris/handle/10665/63821. Accessed on 02.08.2022
18. Bergquist R, Gray DJ. Schistosomiasis elimination: beginning of the end or a continued march on a trodden path. Trop Med Infect Dis. 2019;4(2):76. https://doi.org/10.3390/tropicalmed4020076.
19. Tchuem Tchuenté L-A, Momo SC, Stothard JR, Rollinson D. Efficacy of praziquantel and reinfection patterns in single and mixed infection foci for intestinal and urogenital schistosomiasis in cameroon. Acta Trop. 2013;128:275–83. https://doi.org/10.1016/j.actatropica.2013.06.007.
20. Ndukwe YE, Obiezue RNN, Aguzie ION, Anunobi JT, Okafor FC. Mapping of urinary schistosomiasis in Anambra State Nigeria. Ann Glob Health. 2019;85(1):52. https://doi.org/10.5334/aogh.2393.
21. Pennance T, Person B, Muhsin MA, Khamis AN, Muhsin J, Khamis IS, et al. Urogenital schistosomiasis transmission on Unguja Island, Zanzibar: characterisation of persistent hot-spots. Parasit Vectors. 2016;9:646. https://doi.org/10.1186/s13071-016-1847-0.
22. Rudge JW, Stothard JR, Basanez MG, Mgeni AF, Khamis IS, Khamis AN, et al. Micro-epidemiology of urinary schistosomiasis in Zanzibar: local risk factors associated with distribution of infections among schoolchildren and relevance for control. Acta Trop. 2008;105:45–54. https://doi.org/10.1016/j.actatropica.2007.09.006.
23. Lo NC, Gurarie D, Yoon N, Coulibaly JT, Bendavid E, Andrews JR, et al. Impact and cost-effectiveness of snail control to achieve disease control targets for schistosomiasis. Proc Natl Acad Sci USA. 2018;115:E584–91. https://doi.org/10.1073/pnas.1708729114.
24. King CH, Sutherland LJ, Bertsch D. Systematic review and meta-analysis of the impact of chemical-based mollusciciding for control of Schistosoma mansoni and *S. haematobium* transmission. PLoS Negl Trop Dis. 2015;9:e0004290. https://doi.org/10.1371/journal.pntd.0004290.
25. Li EY, Gurarie D, Lo NC, Zhu X, King CH. Improving public health control of schistosomiasis with a modified WHO strategy: a model-based

comparison study. Lancet Glob Health. 2019;7:e1414–22. https://doi.org/10.1016/S2214-109X(19)30346-8.
26. WHO Regional Office for the Eastern Mediterranean. Report on the intercountry meeting on strategies to eliminate schistosomiasis from the Eastern Mediterranean Region, Muscat, Oman, 6-8 November 2007. 2009. https://apps.who.int/iris/handle/10665/115981. Accessed.on 02.08.2022
27. Madsen H, Rohde R, Maiga AS. Trials on focal molluscicide application in larger irrigation canals and lakes in Mali. Trop Med Parasitol. 1986;37:22–4.
28. Roberts KW, Smith Gueye C, Baltzell K, Ntuku H, McCreesh P, Maglior A, et al. Community acceptance of reactive focal mass drug administration and reactive focal vector control using indoor residual spraying, a mixed-methods study in Zambezi region, Namibia. Malar J. 2021;20:162. https://doi.org/10.1186/s12936-021-03679-1.
29. Hsiang MS, Ntuku H, Roberts KW, Dufour MK, Whittemore B, Tambo M, et al. Effectiveness of reactive focal mass drug administration and reactive focal vector control to reduce malaria transmission in the low malaria-endemic setting of Namibia: a cluster-randomised controlled, open-label, two-by-two factorial design trial. Lancet. 2020;395:1361–73. https://doi.org/10.1016/s0140-6736(20)30470-0.
30. Burnim M, Ivy JA, King CH. Systematic review of community-based, school-based, and combined delivery modes for reaching school-aged children in mass drug administration programs for schistosomiasis. PLoS Negl Trop Dis. 2017;11:e0006043. https://doi.org/10.1371/journal.pntd.0006043.
31. De Rosa E, Person B, Knopp S, Muhsin J, Lyimo JH, Kabole F, et al. A descriptive qualitative case study of the experiences, perceptions and attitudes of pregnant women on Unguja island, Zanzibar, towards antischistosomal treatment. Acta Trop. 2022;225:106143. https://doi.org/10.1016/j.actatropica.2021.106143.
32. Ajibola O, Gulumbe BH, Eze AA, Obishakin E. Tools for detection of schistosomiasis in resource limited settings. Med Sci. 2018;6:39. https://doi.org/10.3390/medsci6020039.
33. Osei E, Nkambule SJ, Vezi PN, Mashamba-Thompson TP. Systematic review and meta-analysis of the diagnostic accuracy of mobile-linked point-of-care diagnostics in sub-Saharan Africa. Diagnostics. 2021;11:1081. https://doi.org/10.3390/diagnostics11061081.
34. Amoah AS, Hoekstra PT, Casacuberta-Partal M, Coffeng LE, Corstjens P, Greco B, et al. Sensitive diagnostic tools and targeted drug administration strategies are needed to eliminate schistosomiasis. Lancet Infect Dis. 2020;20:e165–72. https://doi.org/10.1016/s1473-3099(20)30254-1.
35. Krauth SJ, Greter H, Stete K, Coulibaly JT, Traoré SI, Ngandolo BN, et al. All that is blood is not schistosomiasis: experiences with reagent strip testing for urogenital schistosomiasis with special consideration to very-low prevalence settings. Parasit Vectors. 2015;8:584. https://doi.org/10.1186/s13071-015-1165-y.
36. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Urogenital schistosomiasis elimination in Zanzibar: accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections. Parasit Vectors. 2018;11:552. https://doi.org/10.1186/s13071-018-3136-6.
37. Toor J, Coffeng LE, Hamley JID, Fronterre C, Prada JM, Castaño MS, et al. When, who, and how to sample: designing practical surveillance for 7 neglected tropical diseases as we approach elimination. J Infect Dis. 2020;221:S499-s502. https://doi.org/10.1093/infdis/jiaa198.
38. Rostron P, Pennance T, Bakar F, Rollinson D, Knopp S, Allan F, et al. Development of a recombinase polymerase amplification (RPA) fluorescence assay for the detection of *Schistosoma haematobium*. Parasit Vectors. 2019;12:514. https://doi.org/10.1186/s13071-019-3755-6.
39. McCarthy JS, Lustigman S, Yang GJ, Barakat RM, García HH, Sripa B, et al. A research agenda for helminth diseases of humans: diagnostics for control and elimination programmes. PLoS Negl Trop Dis. 2012;6:1601. https://doi.org/10.1371/journal.pntd.0001601.
40. Gass K. Time for a diagnostic sea-change: Rethinking neglected tropical disease diagnostics to achieve elimination. PLoS Negl Trop Dis. 2020;14:e0008933-e. https://doi.org/10.1371/journal.pntd.0008933.

Publisher's Note

3.6. Test-Treat-Track-Test-Treat (5T) approach for *Schistosoma haematobium* elimination on Pemba Island, Tanzania

Lydia Trippler[1,2], Lyndsay Taylor[1,2], Mohammed Nassor Ali[3], Sarah Omar Najim[3], Khamis Seif Khamis[3], Jan Hattendorf[1,2], Saleh Juma[4], Shaali Makame Ame[5], Fatma Kabole[5], Said Mohammed Ali[3], Stefanie Knopp[1,2]

1 Swiss Tropical and Public Health Institute, Allschwil, Switzerland

2 University of Basel, Basel, Switzerland

3 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

4 Neglected Diseases Program, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania

5 Neglected Diseases Program, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

This article was published in:

BMC Infectious Diseases (2024), Jul 02; 24(1):661

doi: 10.1186/s12879-024-09549-w

BMC Infectious Diseases

RESEARCH Open Access

Test-treat-track-test-treat (5T) approach for *Schistosoma haematobium* elimination on Pemba Island, Tanzania

Lydia Trippler[1,2], Lyndsay Taylor[1,2], Mohammed Nassor Ali[3], Sarah Omar Najim[3], Khamis Seif Khamis[3], Jan Hattendorf[1,2], Saleh Juma[4], Shaali Makame Ame[5], Fatma Kabole[5], Said Mohammed Ali[3] and Stefanie Knopp[1,2*]

Abstract

Background After decades of praziquantel mass drug administration (MDA), several countries approach schistosomiasis elimination. Continuing MDA in largely uninfected populations no longer seems justified. Alternative interventions to maintain the gains or accelerate interruption of transmission are needed. We report results, strengths, and shortcomings of novel test-treat-track-test-treat (5T) interventions in low *Schistosoma haematobium* prevalence areas on Pemba, Tanzania.

Methods School- and household-based surveys were conducted in 2021 and 2022 to monitor the *S. haematobium* and microhematuria prevalence and assess the impact of interventions. In 2021, 5T interventions were implemented in 15 low-prevalence areas and included: (i) testing schoolchildren in primary and Islamic schools for microhematuria as a proxy for *S. haematobium*, (ii) treating positive children, (iii) tracking them to their households and to water bodies they frequented, (iv) testing individuals at households and water bodies, and (v) treating positive individuals. Additionally, test-and-treat interventions were implemented in the 22 health facilities of the study area.

Results The *S. haematobium* prevalence in the school-based survey in 15 low-prevalence implementation units was 0.5% (7/1560) in 2021 and 0.4% (6/1645) in 2022. In the household-based survey, 0.5% (14/2975) and 0.7% (19/2920) of participants were infected with *S. haematobium* in 2021 and 2022, respectively. The microhematuria prevalence, excluding trace results, in the school-based survey was 1.4% (21/1560) in 2021 and 1.5% (24/1645) in 2022. In the household-based survey, it was 3.3% (98/2975) in 2021 and 5.4% (159/2920) in 2022. During the 5T interventions, the microhaematuria prevalence was 3.8% (140/3700) and 5.8% (34/594) in children in primary and Islamic schools, respectively, 17.1% (44/258) in household members, and 16.7% (10/60) in people at water bodies. In health facilities, 19.8% (70/354) of patients tested microhematuria-positive.

Conclusions The targeted 5T interventions maintained the very low *S. haematobium* prevalence and proved straightforward and feasible to identify and treat many of the few *S. haematobium*-infected individuals. Future research will show whether 5T interventions can maintain gains in the longer-term and expedite elimination.

*Correspondence:
Stefanie Knopp
s.knopp@swisstph.ch

Full list of author information is available at the end of the article

Trial registration ISRCTN, ISCRCTN91431493. Registered 11 February 2020, https://www.isrctn.com/ISRCTN91431493.

Keywords Interventions, Case finding, Elimination, Interruption of transmission, Mass drug administration, Schistosomiasis, *S. haematobium*, Surveillance-response, Test-and-treat, Test-treat-track-test-treat, Zanzibar

Background

Human schistosomiasis is a neglected tropical disease (NTD) that can cause serious health consequences. Currently, people from 78 countries are at risk of an infection with *Schistosoma* spp., primarily in sub-Saharan Africa [1, 2]. The primary intervention against schistosomiasis in endemic countries is preventive chemotherapy through mass drug administration (MDA) of praziquantel, implemented by national schistosomiasis control programs [1, 3]. Regular MDA is likely the main driver for the significant reduction in prevalence in sub-Saharan Africa over the past 20 years [4].

Several countries, including Egypt and Morocco [5–8], and certain regions in sub-Saharan Africa, for example in Cameroon [9] and Côte d'Ivoire [10] have now reached very low levels of schistosomiasis and are approaching elimination. Large-scale MDA no longer seems justified in these areas as the majority of the population is not infected with *Schistosoma* spp [11, 12]. Providing praziquantel to uninfected people would be a misuse of resources that are urgently needed elsewhere, specifically in regions where children and adults still suffer from high morbidity. Yet, there is a lack of clear guidelines and evidence at what prevalence levels it is safe to stop praziquantel MDA and what interventions can be used as alternatives. Such alternative interventions should ideally not only sustain the gains made by multiple rounds of MDA and prevent resurgence, but support countries to progress towards the ultimate goal: interruption of transmission.

To reach interruption of transmission, current WHO guidelines recommend that in communities where the prevalence of schistosomiasis is <10%, programs either continue with MDA at the same or reduced frequency or use a clinical approach of test-and-treat if there had not been a program of regular preventive chemotherapy before [3]. However, these recommendations are conditional since the certainty of evidence is still very low [3]. More studies need to be conducted to show if MDA at reduced frequency or test-and-treat approaches indeed can support the elimination goals.

On the Zanzibar islands, Unguja and Pemba, urogenital schistosomiasis caused by infection with *Schistosoma haematobium* used to be a considerable public health problem [11]. However, small and large-scale interventions against schistosomiasis have been implemented for almost 100 years and contributed to a substantial decrease in the prevalence and morbidity [11]. Since 2012, the overall *S. haematobium* prevalence in Zanzibar is below 10% [13–15]. To reach elimination, annual or biannual MDA with praziquantel for children and adults is implemented in schools and communities across the islands since 2013 [13–15]. Additionally, snail control and behavior change communication, are applied in large-scale operational research projects, such as the Zanzibar Elimination of Schistosomiasis project (2012–2017) and the SchistoBreak study (2020–2024) [16, 17]. Since 2017, the islands have reached 'elimination as a public health problem' and aim to proceed towards 'interruption of *Schistosoma* transmission' [11, 13, 18]. In 2020, the overall *S. haematobium* prevalence in Zanzibar was 3.4% in schoolchildren and 0.4% in adults [15]. However, despite the very low overall prevalence, there is a marked heterogeneity of transmission and infection across each island, with many low-prevalence and a few moderate to high prevalence administrative areas [15]. The Zanzibar islands are now facing the challenges of how to address this heterogeneity, of whether, when, and where to stop MDA and of what interventions to use as alternatives to large-scale MDA to maintain the gains or to reach interruption of transmission.

The SchistoBreak study, implemented from 2020 to 2024 in the north of Pemba, aims to address this spatial heterogeneity by targeting different sets of elimination interventions to the local micro-epidemiology of *S. haematobium* [17]. In low-prevalence areas, a surveillance-response approach is implemented as an alternative intervention to MDA. Part of this surveillance-response approach are targeted test-treat-track-test-treat (5T) interventions, where students are tested for urogenital schistosomiasis, treated if positive, and tracked to their homes and water bodies, where household members and people at water bodies, respectively, are also offered testing and treatment if positive. Here, we aimed to assess whether 5T interventions can maintain or further reduce the *S. haematobium* prevalence in low-prevalence areas and report strengths and shortcomings from the first period of targeted 5T interventions implemented in 15 low-prevalence areas in 2021.

Methods

Study setting

The SchistoBreak study is conducted on Pemba, one of the two main islands of the Zanzibar archipelago, United Republic of Tanzania. Pemba is located 30 km off the east coast of the country's mainland. The island is divided

into four districts, "Mkoani", "Chake Chake", "Wete", and "Micheweni", which are subdivided into 129 small administrative areas known as "shehias" [19]. The SchistoBreak study area consists of 20 shehias, which are referred to in this publication as "implementation units" (IUs). The 20 IUs are located in the two northern districts of Pemba, Wete and Micheweni, which are characterized by their mostly rural environment and the presence of numerous water bodies [18]. Recent data from a national census conducted in 2022 indicate a total population of 543,441 for Pemba, with 272,091 people living in the districts Wete and Micheweni [20]. The vast majority of Pemba's population is of the Islamic faith, and it is common for children to attend madrassas (Islamic schools) in the afternoons or on weekends, in addition to attending primary or secondary school. There are 47 health facilities located in the districts Wete and Micheweni, including primary health care units (PHCUs) and hospitals. Of these, 22 are located within the SchistoBreak study area [20]. On Pemba, only urogenital schistosomiasis caused by *Schistosoma haematobium* is endemic, while autochthonously acquired intestinal schistosomiasis is absent [11].

Study design and participants

The SchistoBreak study follows a longitudinal design with three annual intervention periods and four annual school-based and household-based surveys that employ a cross-sectional sampling approach to monitor the *S. haematobium* prevalence in the IUs and to measure the impact of the interventions [17]. The study runs over four years, from February 2020 to March 2024. Annual surveys are conducted from November to February/March each year, before and after the intervention periods from May to October [17]. Different sets of interventions targeted to the local micro-epidemiology are employed. In IUs with a *S. haematobium* prevalence of ≥3% in schoolchildren and ≥2% in communities, a combination of annual MDA, snail control, and behavior change interventions is implemented to accelerate the interruption of *S. haematobium* transmission in "hotspots". In IUs with a *S. haematobium* prevalence of <3% in schoolchildren and <2% in communities, no MDA is provided. Instead, surveillance-response interventions, including the 5T interventions described here, are implemented in schools, households, and at water bodies as described below. Furthermore, in health facilities within all IUs in the study area a simple test-and-treat approach is conducted.

Any person aged≥4 years old enrolled in one of the study schools/madrassas or living in the study area is eligible to participate in the annual surveys and the interventions. Children<4 years old are excluded from the study as it is challenging to collect urine samples from babies and toddlers and as the safety of praziquantel (BILTRICIDE®) has been established only for individuals aged≥4 years [21].

School-based and household-based surveys

From November 2020 to February 2021 and from November 2021 to March 2022, school-based and household-based surveys were implemented in the 20 IUs of the study site. The school-based surveys were conducted in each largest public primary school within the 20 IUs, and the household-based surveys were conducted in each community within the 20 IUs. No school-based survey was conducted if an IU did not have a public primary school.

In each study school, 175 children from nursery school through grade six were randomly selected to participate. The random sampling is described in detail elsewhere [18]. Briefly, a total of 25 children were randomly selected from each grade (nursery school to grade six), balanced by sex. Subsequently, the selected children were registered with demographic details and given consent and assent forms. On the following day, upon return of the signed consent and assent forms, the present children were given a transparent plastic container (100 ml) labeled with a unique identifier code and asked to provide a fresh urine sample.

In each study community, 70 housing structures (in 2021) or 80 housing structures (in 2022), respectively, were randomly preselected. The process used to randomize and locate the housing structures within the communities is described elsewhere in detail [18, 22]. Briefly, a navigation system installed on mobile devices was used to locate each housing structure in the community. If a housing structure was inhabited, after consenting, an adult person living in the house was invited to participate in a questionnaire interview and to disclose sociodemographic details of all household members. Furthermore, all eligible household members were invited to participate in the study by signing an informed consent form and providing a urine sample for examination. Urine samples were provided either immediately at the first visit, or in case of absent household members, until the second visit on the following morning or, if still absent, until the third visit on the third day.

Test-treat-track-test-treat interventions

The first period of 5T interventions was implemented from June 2021 to October 2021 in 15 IUs that had met the criteria of a *S. haematobium* prevalence of ≤3% in the school-based survey and of ≤2% in the household-based survey in the baseline survey in 2021 [17, 18]. The 5T interventions consist of five (5) parts (testing, treating, tracking, testing, and treating). The first and second parts (test, treat) were implemented in schools and madrassas. Since children enrolled in grades 3–5 in Zanzibar are

at the highest risk of infection [15], the initial testing to identify as many positives as possible involved all children in grades 3–5 in one primary school and all children in one madrassa, respectively, per low-prevalence IU. No primary school was located in four of the 15 IUs. Here, only children from madrassas were tested. Before testing, all children were registered with demographic details. In madrassas, children were additionally asked which primary/secondary school they attended, in which grade they were enrolled, and whether they had already been tested (and treated) within the same 5T intervention period to avoid double testing and treating. The following day, once children had submitted the signed consent and assent forms and produced their urine sample between 9 am and 4 pm (depending on school times), the samples were tested for microhematuria as a proxy for *S. haematobium* infection at the point-of-care directly in the schools by the trained field enumerators, using Hemastix reagent strips (Hemastix; Siemens Healthcare Diagnostics AG; Zurich, Switzerland). In the second part (treat), all children who tested microhematuria-positive were treated with a single dose of praziquantel (40 mg/kg body weight) by a member of the Zanzibar Neglected Diseases Program, who was part of our research team, using a dose-pole [23]. Next, in the third part (track), all positive-tested children were accompanied to their homes and to water bodies they frequently used. Water bodies located outside the study area were not visited. Our assumption was that household members of positive children may use the same water bodies where transmission occurs and, as other people that use these water bodies, be at high risk of infection. In the fourth and fifth parts (test, treat), all individuals present at the time of the visit at the homes and the water bodies were invited to respond to demographic questions and to provide a fresh urine sample after written consent. Both households and water bodies were visited once. The study team spend up to two hours at each water body to invite present or arriving individuals to test and treat activities. Each urine sample was tested for microhematuria on site by the trained field enumerators and subsequently taken to the Public Health Laboratory – Ivo de Carneri (PHL-IdC) in Chake Chake, Pemba, for *S. haematobium* egg detection (see below). If a household member or individual present at a water body tested positive for microhematuria and/or *S. haematobium* eggs, the individual was offered immediate treatment with praziquantel (40 mg/kg body weight) on site (if positive for microhematuria) or called by phone and encouraged to seek treatment at the nearest health facility (if positive for *S. haematobium* eggs by urine filtration).

Test-and-treat in health facilities

In June 2021, staff from each of the 22 health facilities (20 PHCUs and two hospitals) in the study area were invited to the PHL-IdC for a meeting, including a presentation about the life cycle, symptoms, and health consequences of urogenital schistosomiasis, discussion, and training on the use of Hemastix reagent strips to test for microhematuria in urine samples. Subsequently, the staff were equipped with Hemastix reagent strips to test for microhematuria in patients presenting with symptoms consistent with urogenital schistosomiasis, such as visible hematuria, painful and/or frequent urination, abdominal pain, or infertility within the routine services of their health facility. They were also equipped with praziquantel to treat individuals who tested microhematuria-positive. In addition, the staff were requested to keep a record of patients presenting with schistosomiasis-related symptoms, were tested for microhematuria and treated with praziquantel (40 mg/kg body weight) if positive. These records were collected every two weeks from July 2021 onwards by a member of the SchistoBreak study team.

Laboratory examinations

All urine samples collected during the surveys and at the households and water bodies during the 5T intervention period were transported to the PHL-IdC. Here, 10 ml of each urine sample were filtered through a 13 mm fabric filter (Sefar Ltd., Bury, UK), which was placed in a Swinnex plastic filter holder (Millipore-Merck KGaA, Darmstadt, Germany) attached to a plastic syringe. The filters were placed on a microscope slide labeled with the participant's unique identifier code and examined under a light microscope for the presence and number of *S. haematobium* eggs. In addition, all urine samples collected during the surveys were examined for microhematuria on the same day using Hemastix reagent strips. All examinations in the laboratory were conducted by three experienced laboratory technicians who recorded the results for each identifier code on case report forms. As the urine samples collected during the 5T interventions were already tested for microhematuria at the point-of-care in the field by trained field enumerators, the testing was not repeated in the laboratory.

Data management

All demographic data collected in the school-based survey and all data collected during the questionnaire interviews in the household-based survey were captured by inserting responses into the preprogrammed questionnaire application Open Data Kit (ODK, www.opendatakit.org), installed on Samsung Galaxy Tab A tablets. All data containing results of Hemastix reagent strip examinations conducted at the point-of-care in schools, households, or at water bodies during the 5T intervention

period were also captured through ODK. These electronic data were subsequently sent to the ODK server, hosted by the Swiss Tropical and Public Health Institute (Swiss TPH) in Allschwil, Switzerland. All results of laboratory examinations of urine samples, including the results of urine filtrations and Hemastix reagent strips collected during the surveys, were recorded on paper and then double-entered into a Microsoft Excel spreadsheet (2016, version 16.0.5395.1000) by two experienced data entry clerks from PHL-IdC. These spreadsheets were sent to the Swiss TPH via a secure server and cleaned using R version 4.0.3 (www.r-project.org) and Stata/IC 16 (StataCorp LLC, College Station, TX, USA). Mismatching double-entry results were checked against the original paper forms and reentered correctly. For statistical analysis, the laboratory results were merged with the questionnaire and registration data based on the unique identifier codes. To inform participants about their *S. haematobium* infection status, participant names were merged once with laboratory analysis data. Otherwise, names were kept in a separate file, and only coded data were used for statistical analyses.

Statistical methods

Statistical analyses for this publication were conducted using R version 4.1.3.

Only data collected in IUs classified as low-prevalence IUs during the baseline survey of the SchistoBreak study [18] were included in the analysis of this publication. One exception are the data collected in the 22 health facilities that were spread across the study area. All data from patients in health facilities were included in the analysis when patients were tested for microhematuria between July 2021 (the start of the collaboration with health facilities) and November 2021 (the end of the first intervention period).

All eligible individuals who provided a urine sample were included in the parasitological analyses of the surveys, the 5T interventions, and the test-and-treat interventions in health facilities. A participant was considered microhematuria-positive if the result of Hemastix reagent strips was trace, small (+), moderate (++), or large (+++) according to the color chart provided by the manufacturer. A participant was considered positive for *S. haematobium* if one or more eggs were detected by urine filtration.

Prevalence of microhematuria and *S. haematobium* in the surveys, 5T interventions, and test-and-treat interventions in health facilities was calculated overall and per IU for each survey or intervention period, respectively. The statistical significant difference of prevalence between the survey years was calculated for each survey using a two-sample proportion test.

For children tested in madrassas, the children's potential additional enrolment in a primary or secondary school and their grade level were assessed to determine how many children were captured in madrassas but were not part of the 5T interventions in primary schools. Moreover, the microhaematuria prevalence of madrassa students that also attended a public school *versus* non-school attendees was determined.

The sites of data collection during the 5T interventions, including screened schools and madrassas and tracked households and water bodies, were mapped with ArcMap version 10.6.1 (ESRI, Redlands, CA, USA). The locations of households were geographically masked in order to preserve confidentiality.

Results

Participation in school-based and household-based surveys 2021 and 2022

The number of children and household members enrolled and the overall results of the baseline school-based and household-based survey, respectively, in all 20 IUs carried out from November 2020 to February 2021 are presented elsewhere [18]. This baseline survey revealed a total of five hotspot IUs according to the preset prevalence threshold criteria. Additional 15 IUs were identified as low-prevalence, including 11 low-prevalence primary schools (no primary school was located in four IUs) and 15 low-prevalence communities. Subsequent results focus on these 11 low-prevalence primary schools and 15 low-prevalence communities, since the 5T interventions that are the primary focus of this publication were only implemented in the low-prevalence IUs.

In the school-based survey conducted in 2021, a total of 1924 children were registered across the 11 low-prevalence schools. Of these 1924 children, 332 (17.3%) were absent on the day of urine collection, and 32 (1.7%) refused to participate or did not provide a signed consent form, resulting in 1560 (81.1%) children providing a fresh urine sample for examination (Supplementary File 1: Fig. 1A).

In the household-based survey in 2021, 3539 household members were registered across the 15 low-prevalence communities. Of these 3539 household members, 564 (15.9%) were absent on the day of urine collection, refused to participate, or did not provide a signed consent form. Hence, a total of 2975 (84.1%) household members provided a fresh urine sample for examination (Supplementary File 1: Fig. 1A).

In the school-based survey in 2022, a total of 1898 children were registered across the 11 low-prevalence schools. Of these 1898 children, 238 (12.5%) were absent on the day of urine collection, and 15 (0.8%) refused to participate or did not provide a signed consent form.

Hence, 1645 (86.7%) children provided a fresh urine sample for examination (Supplementary File 1: Fig. 1B).

In the household-based survey in 2022, 3683 household members were registered across the 15 low-prevalence communities. Of these 3683 household members, one (0.03%) person was excluded due to ineligibility (age < 4 years), 755 (20.5%) household members were absent on the day of urine collection, and 7 (0.2%) individuals refused to participate or did not provide a signed consent form, resulting in 2920 (79.3%) household members providing a fresh urine sample (Supplementary File 1: Fig. 1B).

Demographic information from participants of all surveys and interventions is shown in Table 1.

Participation in the 5T intervention period in 2021

Between May and October 2021, a total of 4283 and 777 children were registered in 11 schools and 15 madrassas, respectively, across the 15 low-prevalence IUs (Fig. 1). Of the 4283 children registered in the primary schools, 540 (12.6%) were absent on the day of urine collection and 39 (0.9%) refused to participate or did not provide a signed consent form. Hence, 3704 (86.5%) children were tested for microhematuria and included in the analysis. Of the 777 children registered in the madrassas, 164 (21.1%) were absent on the day of urine collection, and 19 (2.4%) refused to participate or did not provide a signed consent form. Hence, a total of 594 (76.4%) children were tested for microhematuria and included in the analysis.

Of the 3704 children tested in primary schools and 594 children tested in madrassas, 190 (5.1%) and 47 (7.9%), respectively, were microhematuria-positive.

Of the 237 microhematuria-positive children, 215 (90.7%) were tracked to 200 households. In 89 (44.5%) of the 200 households, nobody was present at the time of visit or all present members refused to participate. In the remaining 111 households, of the 280 household members present at the time of the visit, 22 (7.9%) refused to participate or did not provide a signed consent form. A total of 258 (92.1%) household members provided a fresh urine sample and had demographic data recorded.

Of the 237 microhematuria-positive children, 189 (79.7%) were tracked to 61 water bodies. At 15 water bodies, a total of 61 individuals were present at the time of the visit. Of the 61 individuals, one (1.6%) person refused to participate or did not provide a signed consent form. A total of 60 (98.4%) individuals provided a fresh urine sample and had demographic data recorded.

Information about sex and median age of the study participants, stratified per survey and intervention period, is indicated in Table 1.

Table 1 Demographic information of study participants. Demographic information of participants in school-based and household-based surveys and test-treat-track-test-treat (5T) activities implemented in 15 low *Schistosoma haematobium* prevalence implementation units in the North of Pemba, Tanzania, in 2021–2022

		2021 Parasitological survey			2021 Test-Treat-Track-Test-Treat intervention period							2022 Parasitological survey		
		2021 school-based survey		2021 household-based survey		2021 primary school testing		2021 madrassa testing		2021 household tracking		2021 water body tracking	2021 health facilities test-and-treat		2022 school-based survey		2022 household-based survey	
N		1560		2975		3704		594		258		60	354		1645		2920	
	Female n (%)	839	(53.8)	1570	(52.8)	1942	(52.4)	284	(47.8)	152	(58.9)	27 (45.0)	201	(56.8)	855	(52.0)	1617	(55.4)
	Male n (%)	721	(46.2)	1405	(47.2)	1762	(47.6)	310	(52.2)	106	(41.1)	33 (55.0)	153	(43.2)	790	(48.0)	1303	(44.6)
Age (median)		10		17		11		9		16.5		13	20		10		18	

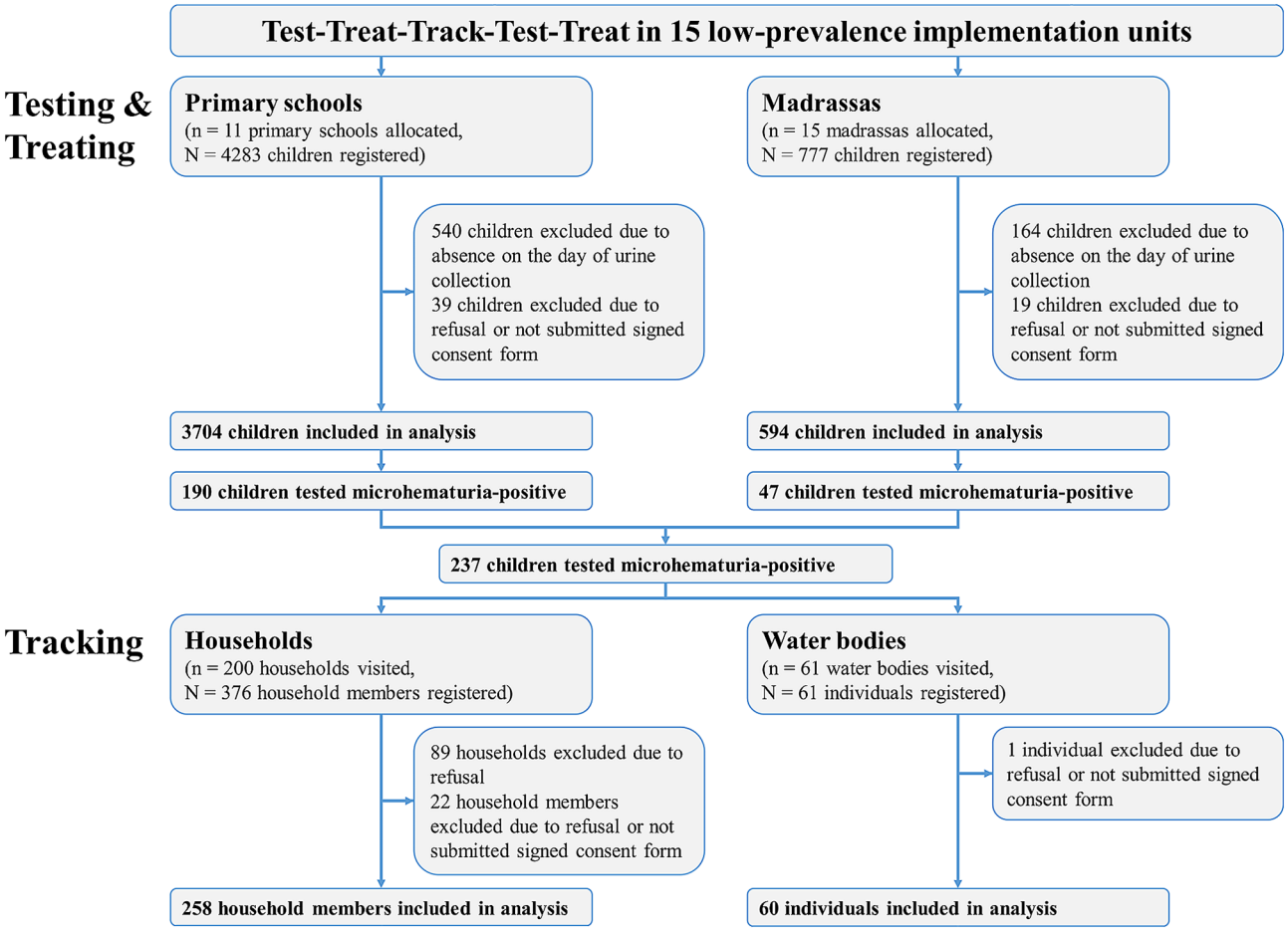


Fig. 1 Participation in test-treat-track-test-treat interventions. Flow diagram of individuals participating in test-treat-track-test-treat interventions in 15 low *Schistosoma haematobium* prevalence implementation units in the North of Pemba, Tanzania, in 2021

Microhematuria and *Schistosoma haematobium* prevalence in surveys, test-treat-track-test-treat intervention period, and health facilities

The overall prevalence of microhematuria among children who participated in the school-based surveys in the 15 low-prevalence IUs was 3.1% (47/1533; 95% CI: 2.3–4.1) in 2021 and 6.3% (104/1644; 95% CI: 5.2–7.6) in 2022 (Fig. 2A and Supplementary File 2: Table 1). The results of the two-sample proportion test showed that the prevalence between the two years was significantly different ($p<0.0001$). The prevalence of microhematuria among participants of the household-based survey was 5.5% (162/2970; 95% CI: 4.7–6.4) in 2021 and 13.2% (386/2920; 95% CI: 12.0–14.5) in 2022. The results of the two-sample proportion test showed that the prevalence between the two years was significantly different ($p<0.0001$).

The prevalence of microhematuria excluding trace results among children who participated in the school-based surveys in the 15 low-prevalence IUs was 1.4% (21/1533; 95% CI: 0.9–2.1) in 2021 and 1.5% (24/1644; 95% CI: 1.0–2.2) in 2022. The results of the two-sample proportion test showed that the prevalence between the two years was not significantly different ($p=0.93$). The prevalence of microhematuria among participants of the household-based survey was 3.3% (98/2975; 95% CI: 2.7–4.0) in 2021 and 5.4% (159/2920; 95% CI: 4.6–6.3) in 2022. The results of the two-sample proportion test showed that the prevalence between the two years was significantly different ($p<0.0001$).

Figure 2B shows that the overall *S. haematobium* prevalence, as assessed in the school-based surveys, was 0.5% (7/1560; 95% CI: 0.2–1.0) in 2021 and 0.4% (6/1645; 95% CI: 0.2–0.9) in 2022 in schoolchildren attending the 11 primary schools of the 15 low-prevalence IUs. The results of the two-sample proportion test showed that the prevalence between the two years was not significantly different ($p=0.87$). The *S. haematobium* prevalence, assessed in the household-based survey in the communities of the 15 low-prevalence IUs, was 0.5% (14/2975; 95% CI: 0.3–0.8) in 2021 and 0.7% (19/2920; 95% CI: 0.4–1.1) in 2022. The results of the two-sample proportion test showed that the prevalence between the two years was not significantly different ($p=0.41$).

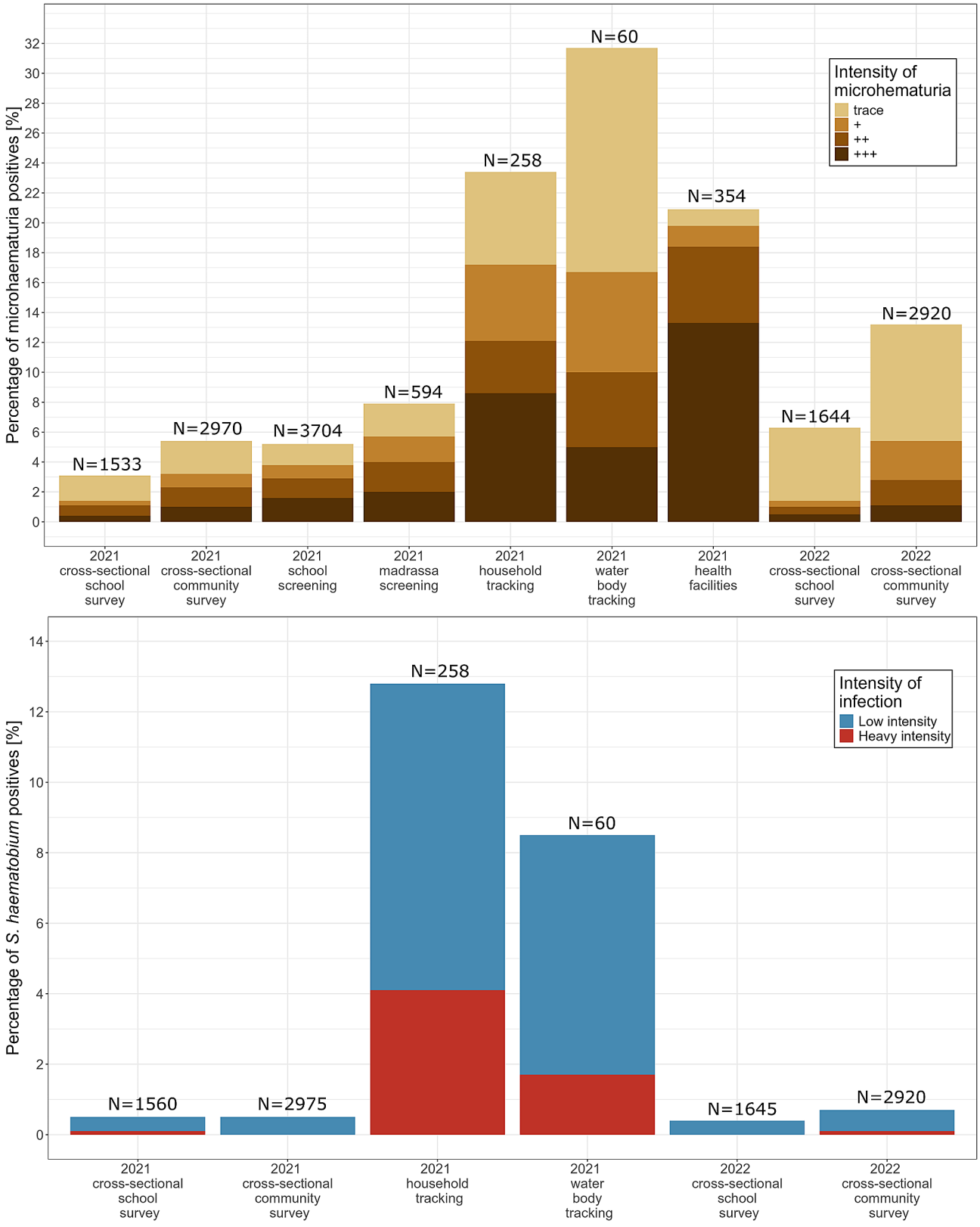


Fig. 2 Microhematuria (**A**) and *Schistosoma haematobium* (**B**) prevalence in 15 low-prevalence implementation units in Pemba, Tanzania, in 2021–2022. The y-axis represents the percentage of positive-tested individuals for microhematuria (**A**) or *S. haematobium* infection (**B**). Colors indicate the grading of microhematuria or intensity of *S. haematobium* infections. N indicates the overall number of tested individuals per bar

During the 5T intervention period in 2021, 5.1% (190/3704) of the children tested in primary schools and 8.0% (47/594) of the children tested in madrassas were microhematuria-positive (Fig. 2A). Of the 190 and 47 children tested positive for microhematuria in schools and madrassas, 180 (95.0%) and 42 (89.4%), respectively, were treated with praziquantel after testing. In the households and at the water bodies identified by tracking these microhematuria-positive children, 23.3% (60/258) and 31.7% (19/60), respectively, of the tested individuals were microhematuria-positive. Of the 60 and 19 individuals who tested positive in households and at water bodies, 98.3% (59/60) and 94.7% (18/19), respectively, were treated at the point-of-care.

The risk-based tracking of microhematuria-positive children from primary schools or madrassas to their homes identified 12.8% (31/243) of household members present at home at the time of the visit as *S. haematobium* egg-positive (Fig. 2B). At the water bodies that were identified by tracking microhematuria-positive children, 8.5% (5/60) of individuals present at the water bodies at the time of the visit were tested *S. haematobium* egg-positive.

Between August and November 2021, 354 patients presented with symptoms aligning with urogenital schistosomiasis in 13 of the 22 health facilities in the study area. All 354 individuals were tested with Hemastix reagent strips at the point-of-care. Among them, 20.9% (74/354) were microhematuria-positive (Fig. 2A). Of these 74 individuals, 90.5% (67/74) were treated with praziquantel in the health facilities.

Information on microhematuria and *S. haematobium* prevalence for each of the surveyed, tested and tracked populations, stratified by sex, are presented in Supplementary File 2: Table 1.

Heterogeneity of microhematuria and *S. haematobium* prevalence during the test-treat-track-test-treat intervention period

The testing during the 5T intervention period showed a varying microhematuria prevalence between 0.0% and 15.6% in schools and between 0.0% and 30.0% in madrassas (Fig. 3A). The school and madrassa with the highest microhematuria prevalence were located in the same IU. During the risk-based tracking of children who tested positive for microhematuria in schools and madrassas, the microhematuria prevalence per IU varied between 0.0% and 47.6% in households and between 0.0% and 56.2% at water bodies, respectively.

In the 5T intervention period, the *S. haematobium* prevalence per IU ranged from 0.0 to 40.0% in households and from 0.0 to 25.0% at the water bodies, respectively (Fig. 3B).

In the test-and-treat intervention in the health facilities, the microhematuria prevalence ranged from 0.0 to 100% in health facilities (Fig. 3A). However, it should be noted that all health facilities with a microhematuria prevalence of 100% had tested < 5 patients with urogenital schistosomiasis-related symptoms.

Children tested in madrassas during the test-treat-track-test-treat activities

Of the 594 madrassa students who were tested for microhematuria, 219 (36.9%) attended a primary school that was part of our 5T interventions, but were enrolled in grades where screening for microhematuria was not operated, i.e., in nursery school, grades 1, 2, or grade 6. Another 79 (31.3%) of the 594 tested madrassa students reported that they would not visit a school. For 132 (22.2%) children, no information was available on their primary/secondary school enrolment and attendance because the assessment of primary school enrolment was only started from the third madrassa onwards. Another 97 (16.3%) of the 594 madrassa students were enrolled in primary schools that were not part of the SchistoBreak study schools. A total of 51 (8.6%) children were enrolled in grades 3–5 in one of the SchistoBreak study schools but were not present on the testing days, and 16 (2.7%) children were enrolled in secondary schools, which were not part of our 5T interventions. Of the 79 non-school attendees, 5 (6.3%) children were tested microhaematuria-positive. The microhaematuria prevalence in school attendees tested in madrassas was 9.1% (35/383).

Households and water bodies identified during the test-treat-track-test-treat interventions

The households of microhematuria-positive children were located between 8 and 4794 m (mean: 958 m) from the school or madrassa where the children were tested. Of the 215 children who were tracked to their homes, 71 (32.0%) children lived in households not located in the same IU as the school the children were attending (Fig. 4).

The water bodies indicated by microhematuria-positive children were located between 62 m and 10.1 km (mean: 1210 m) away from the school or madrassa where the children were tested. For 27.2% (43/158) of the positive-tested children, the water bodies they frequented were not located in the same IU as the screened school. Moreover, several water bodies were located at borders between two IUs and borders to shehias outside of the study area.

In two schools and five madrassas, respectively, no child tested positive for microhematuria, and therefore, no child was tracked to household or water bodies.

Fig. 3 Microhematuria (**A**) and *Schistosoma haematobium* (**B**) prevalence in 15 low-prevalence implementation units in Pemba, Tanzania, in 2021. The y-axis represents the prevalence of microhematuria (**A**) or *S. haematobium* infection (**B**); each point represents one school/community. Colors indicate the grading of microhematuria or *S. haematobium* intensity infections, respectively, per school/community. The asterisk (*) denotes points where the prevalence was calculated with < 5 participants per school/community

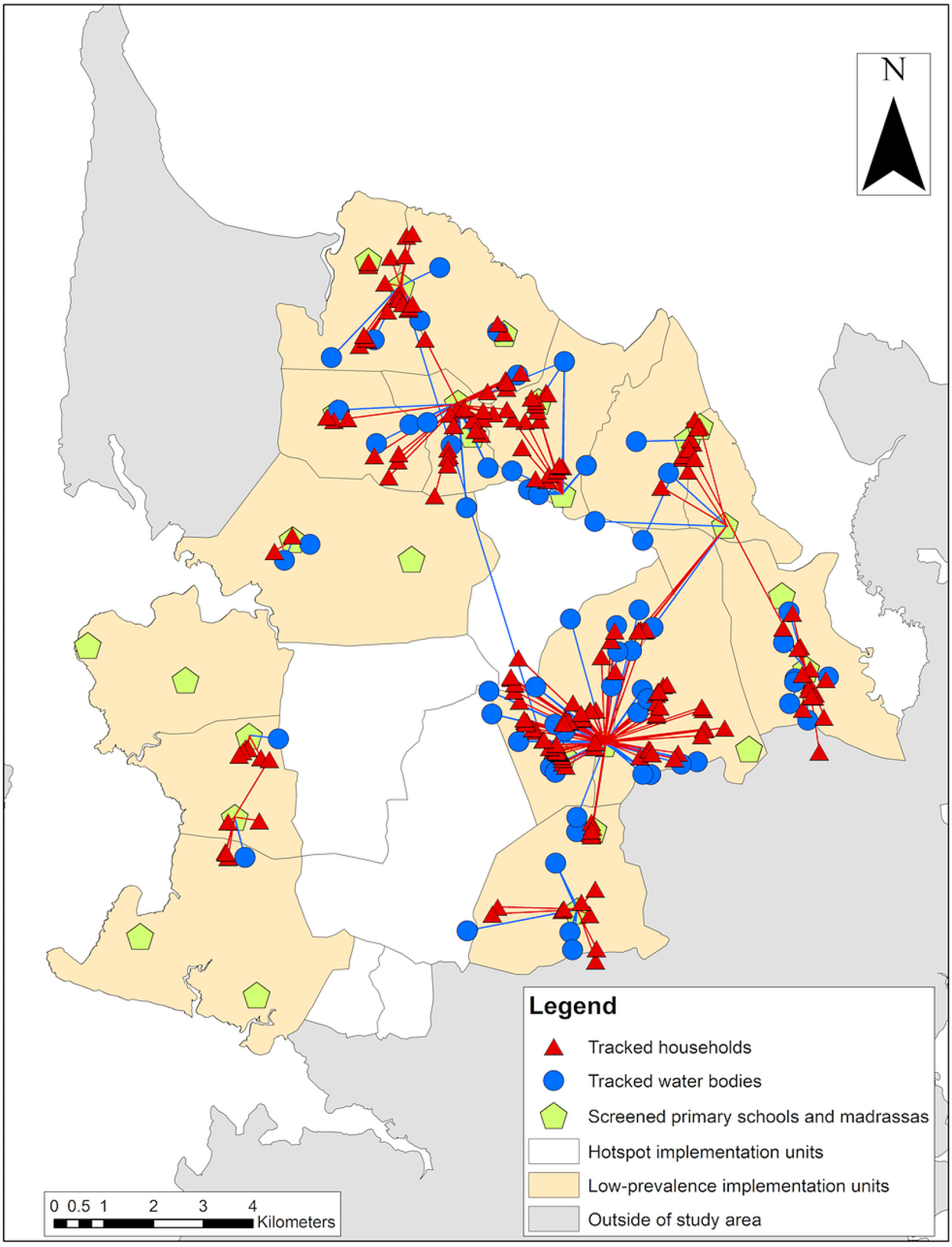


Fig. 4 Spatial distribution of tracked households and water bodies. Spatial distribution of households of microhematuria-positive children and water bodies used and indicated by microhematuria-positive children emanating from the school/madrassa where the children were tested in the 15 low *Schistosoma haematobium* prevalence implementation units in the North of Pemba, Tanzania, in 2021. The locations of households are geographically masked in order to preserve confidentiality

Individuals treated and praziquantel tablets used during the mass drugs administration in 2020 and the 5T interventions in 2021

During the MDA in August 2020, 39,312 individuals were treated with praziquantel in the communities and schools of the 15 IUs that were identified as low-prevalence IUs in 2021 (Table 2). To treat these individuals, 78,853 praziquantel tablets were used, with an average of 2 tablets per treated person. During the 5T interventions in 2021, 336 individuals were treated in primary schools, madrassas, at households and at water bodies in the 15 low-prevalence IUs. For the 336 individuals, 1123 praziquantel tablets were used, with an average of 3 tablets per treated person. Hence, during the MDA in August 2020 around 70 times as many praziquantel tablets were used as during the targeted 5T interventions in 2021 in the same IUs. An additional 85 praziquantel tablets were used to treat 31 hematuria-positive individuals in the health facilities across the study area.

Discussion

The Zanzibar islands have achieved the elimination of urogenital schistosomiasis as a public health problem in 2017 and have aimed for interruption of transmission ever since [11, 13]. Considering that in many communities on the islands the majority of people are not infected with *S. haematobium*, large-scale praziquantel MDA seems no longer justified. Alternative intervention strategies that maintain the gains made by MDA over the past decades and ideally allow Zanzibar to accelerate elimination need to be explored.

Over the first year of the SchistoBreak study, we assessed whether targeted 5T interventions were able to maintain or even reduce the prevalence of urogenital schistosomiasis in 15 low-prevalence shehias in the north of Pemba. Our results showed no considerable change in the prevalence of microhematuria (excluding trace results) and of *S. haematobium* infections after one year of 5T interventions from 2021 to 2022. Hence, aligned with targeted snail control measures that were also part of the surveillance-response activities [17], the 5T interventions were able to maintain the very low overall prevalence, and no outbreaks or major recrudescence were observed. However, the prevalence of microhematuria including trace increased significantly from 2021 to 2022 in the school-based and community-based surveys. While this observation warrants close monitoring in the following study years, since increasing (trace) microhematuria might be a first sign of infections and recurrent morbidity in individuals missed by the 5T interventions, our current assumption is that the increase in trace microhematuria was caused by different and newer batches of Hemastix that were started to be used after the baseline survey in 2021. Other studies from Pemba and Tanzania mainland confirm that older/expired strips have a less intense color reaction [24] and recommend that trace results should be considered negative for *S. haematobium* infections when a high specificity is aimed for in settings with low egg counts in urine filtration [25].

With certainty, however, we can say that the 5T interventions did not contribute to further reducing the overall microhematuria or *S. haematobium* prevalence in our study within one year. Whether the 5T approach is an appropriate intervention to accelerate elimination and progress towards interruption of transmission remains in question, and more evidence needs to be generated in

Table 2 Praziquantel tablets administered during mass drug administration (MDA) in August 2020 and during targeted test-and-treat interventions in 2021. Number of praziquantel tablets administered during the MDA in August 2020 and the test-treat-track-test-treat (5T) intervention period from May to October 2021 and number of praziquantel tablets used during test-and-treat activities in health facilities from July to November 2021 in 15 low *Schistosoma haematobium* prevalence implementation units in the north of Pemba, Tanzania. NA = not applicable because the information is missing. "-" indicates target points where MDA was not planned and conducted

District(s)	Location of treatment interventions	Number of praziquantel tablets used in MDA in August 2020	Number of individuals treated in MDA in August 2020	Number of praziquantel tablets used in 5T and test-and-treat interventions in 2021	Number of individuals treated in 5T and test-and-treat interventions in 2021
Wete	Primary schools	6316	3378	239.5	110
	Madrassas	NA	NA	103	53
	Households	11,290	3924	22.5	8
	Water bodies	-	-	10	12
Micheweni	Primary schools	18,718	10,765	544	53
	Madrassas	NA	NA	70.5	43
	Households	42,529	21,245	120.5	51
	Water bodies	-	-	13	6
	Total	**78,853**	**39,312**	**1123**	**336**
Wete and Micheweni	Health facilities	-	-	85	31
	Total	**78,853**	**39,312**	**1208**	**367**

long-term studies. Close monitoring and regular evaluation is crucial once MDA is stopped and alternative targeted interventions are applied, to allow a timely reaction in case of any sign of recrudescence [26].

In our study, the 5T interventions were an excellent and straightforward approach to identifying many of the few *S. haematobium*-infected individuals in schools, in households of microhematuria-positive children, and at water bodies used by these children. Compared with the very low overall prevalence in schools and communities, a percentage of 12.8% of household members that were infected with *S. haematobium* confirmed the assumption that household members of positive children are also likely to be infected since they may use the same water bodies where transmission occurs. Moreover, the freshwater bodies that microhematuria-positive children reported to use were ideal places to identify and treat additional infected individuals: a percentage of 8.5% of people using these water bodies tested positive for *S. haematobium*. Our results are in line with a study that assessed similar 5T interventions for *S. mansoni* control in two villages in mainland Tanzania, which revealed an even higher percentage (46.8%) of *S. mansoni* infections among household members of positive-tested children [27]. Since this study used a prevalence threshold of <10% to define a low-prevalence IU, the research team was likely working in a setting with a considerably higher prevalence than that in Pemba, and the higher percentage of infected household members is not surprising. The study team from mainland Tanzania also tracked friends of *S. mansoni*-positive children and their household members and identified 37.5% of the friends and 47.1% of the friends' household members as infected, showing that testing friends and their close contacts can be another important supplement when recommending 5T interventions for implementation.

In addition to public primary schools, madrassas proved to be useful venues for the initial testing of children. While more than one third of the children who attended our study madrassas were also enrolled in our study primary schools, they attended grades that were not included in our testing. These students could have also been reached through extended testing in primary schools, which we did not have the capacity and resources to do. However, another third of the tested madrassa students did not attend a primary school, either because they were too young or for other unknown reasons. Thus, as suggested for behavior change communication about schistosomiasis [28], madrassas offer a fantastic opportunity to include non-school attendees into 5T interventions who otherwise would not have been reached but who may have a strong exposure to the parasite during their daily activities. The microhematuria prevalence in non-school attendees and school attendees tested in madrassas was 6.3% and 9.1%, respectively. Hence, while non-school attendees seemed not at higher risk for obtaining a *S. haematobium* infection than school attendees in our study, they are yet an important group for inclusion in 5T interventions.

In our study, with a team of four enumerators, more than 4000 children were tested in five months, and 200 microhematuria-positive children were tracked to their households and the water bodies they had used. Since the mean distance between the schools/madrassas and households or frequented water bodies was only about 1000 m, tracking children to these points was mostly easily conducted. A challenge during the 5T interventions, however, was the considerably low number of individuals who were present at the water bodies at times when the study teams visited, in addition to only few individuals who agreed to participate in test-and-treat activities at households and at water bodies. To reach more people with 5T interventions, good outreach, and awareness campaigns to increase health literacy about urogenital schistosomiasis are needed in target areas. Moreover, it may be worth returning to the same water body several times and staying there for an extended period, as was done in a study in Egypt where the enumerators spent 11 consecutive hours at tracked water bodies to offer test-and-treat to present or arriving individuals [12]. Working with a larger team or multiple teams in our study would undoubtedly have allowed us to test an even larger number of children and schools, to visit households and water bodies at more ideal times and for longer hours, to conduct multiple rounds of testing per school, and to follow-up treated individuals in a similar time frame. This would have increased the overall coverage of test-and-treat and improved treatment outcomes and would hence potentially have resulted in better progress towards interruption of transmission. However, more staff and diagnostic material have cost implications and our financial resources were limited.

Test-and-treat interventions were also offered by health facilities in the study area to patients with symptoms consistent with a *S. haematobium* infection. In health facilities, patients were tested exclusively with Hemastix reagent strips, and the very high percentage of microhematuria in the patients (20.9%) compared with microhematuria levels we found in community surveys indicates that health facilities may offer another important location for reaching and treating infected individuals. For this purpose, however, it needs to be ensured that health facilities have testing equipment and can provide access to praziquantel as recommended by WHO [3, 29]. Microhematuria-positive patients from our study health facilities were not tracked, in contrast to another study from Egypt, where health facility patients with *S. mansoni* infection were tracked, and their household members

were tested for schistosomiasis and treated if positive [12]. Also in malaria surveillance research and practice, tracking malaria patients identified in health facilities to their homes and testing and treating household members and surrounding households is a renowned approach [30, 31]. In future schistosomiasis elimination activities, if resources allow, it may be worthwhile to incorporate the tracking, testing, and treating of household members of individuals who tested positive at health facilities to increase the coverage further.

As with MDA and any other intervention, good coverage is key to success. But resources are limited, and careful decisions have to be made about where to save and where to spend to ensure a cost-effective approach. What our study clearly showed, in addition to the other results, was that the number of praziquantel tablets required during the 5T interventions to treat infected individuals in schools, households, water bodies, and health facilities was much lower than the number of praziquantel tablets required during an MDA. These findings suggest that, by avoiding recrudescence and preventing an overtreatment of a mostly healthy population, 5T interventions may be considered a cost-effective and feasible alternative to MDA to maintain current gains in very low-prevalence settings. However, solid evidence on cost-effectiveness yet remains to be generated.

The inexistence of a sensitive and specific point-of-care test for *S. haematobium* diagnosis was a considerable challenge for our 5T approach. We used a combination of reagent strip and urine filtration tests conducted on a single urine sample to allow a maximum sensitivity to ensure that infected individuals were detected and treated. However, since the sensitivity of both tests to detect light intensity infections is low [32–34], several infected individuals may have been missed. Conversely, since the specificity of reagent strips to diagnose urogenital schistosomiasis is also low, especially in low-prevalence settings [33, 35], uninfected individuals may have been treated and tracked. Hence, the timely development of highly sensitive and specific point-of-care tests for the diagnosis of schistosomiasis (and other NTDs) in elimination settings is crucial to focus all efforts on true positives and thus further enhance the cost-effectiveness of the 5T interventions [36, 37].

Conclusions

The 5T interventions described here were able to maintain the very low *S. haematobium* prevalence in our study IUs in the north of Pemba, and no outbreaks or major recrudescence were observed over the first year of implementation. While it remains in question whether it will expedite the interruption of transmission, the 5T interventions proved to be a straightforward and feasible approach to identifying and treating many of the few *S. haematobium*-infected individuals in an elimination setting. In addition to schools, madrassas were an excellent starting point for testing a high-risk population, including non-school attendees, which would then lead us to additional individuals with a high likelihood of exposure and infection in households and at water bodies. Health facilities were also an important venue for testing and treating patients with symptoms of urogenital schistosomiasis and confirmed microhematuria. Essential for a successful and cost-effective implementation of 5T interventions in elimination settings in the future will be the development of a highly sensitive and specific rapid diagnostic test that can be used at the point-of-care and enables a timely and accurate treatment and tracking of infected individuals. Close monitoring of the endemic situation after MDA has been stopped will be key to allow a timely reaction to recrudescence. More long-term and large-scale studies assessing the feasibility, impact and cost-effectiveness of 5T interventions in different prevalence and environmental settings are needed before it can be recommended to schistosomiasis elimination programs as an alternative or successor to MDA. Most importantly, it remains to be clarified whether 5T interventions are indeed capable of maintaining the gains made by control programs over extended periods and whether or not they can support programs that have the ultimate aim to interrupt *Schistosoma* transmission.

Abbreviations

5T	Test-treat-track-test-treat
IU	Implementation unit
MDA	Mass drug administration
NTD	Neglected tropical disease
ODK	Open Data Kit
PHCU	Primary health care unit
PHL-IdC	Public Health Laboratory – Ivo de Carneri
Swiss TPH	Swiss Tropical and Public Health Institute
WHO	World Health Organization

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12879-024-09549-w.

Supplementary Material 1: Figure 1. Flow diagram of individuals participating in the school-based and household-based surveys in low *Schistosoma haematobium* prevalence shehias on Pemba, Tanzania, in 2021 (A) and 2022 (B)

Supplementary Material 2: Table 1. Prevalence and intensity of microhematuria and *Schistosoma haematobium* infections of participants in school-based and household-based surveys 2021 and 2022, and test-treat-track-test-treat (5T) activities in 2021. NA = Not applicable

Acknowledgements

We thank the Zanzibar Ministry of Health, the Shehas (village leaders), the health facility staff, and all the teachers involved in our study for having the courage to break new ground with us for test-and-treat activities, to open the doors and to support the SchistoBreak interventions. Furthermore, we are grateful for the tremendous support of all the field enumerators, laboratory staff, and data entry clerks - testing thousands of study participants and

providing timely results would not be possible without them. We are indebted to all the participants in the surveys and the 5T interventions for their trust and willingness to participate. A particular thanks goes to the Zanzibar Neglected Tropical Diseases Program, which provided us with the praziquantel tablets needed during the 5T interventions.

Author contributions
L.Tr., Sa.M.A., J.H. and S.K. conceptualized the SchistoBreak study and Sa.M.A. and S.K. initiated the SchistoBreak study. L.Tr., L.T.a and S.K. performed the data curation. L.Tr. conducted the formal analysis and data visualization for the research article. S.K. acquired the funding for the SchistoBreak study. M.N.A., S.O.N. and K.S.K. contributed significantly to the data collection and implementation of surveys and the 5T interventions. L.Tr., J.H. and S.K. conceptualized the statistical methods of the study. L.Tr., L.Ta., S.J., Sh.M.A., Sa.M.A., F.K. and S.K. were responsible for the management of the research activity planning. L.Tr., L.Ta., Sh.M.A., Sa.M.A. and S.K. mentored the core team and M.N.A., S.J., Sh.M.A., Sa.M.A. and F.K. advised for local conditions. L.Tr. and S.K. drafted the first version of the research article. All authors have made substantial contributions to the study, and have reviewed and approved the submitted version of the manuscript.

Funding
Funding for the study has been obtained from the Swiss National Science Foundation (SNSF) via the PRIMA grant (PR00P3_179753 / 1) of Stefanie Knopp. This funding source had no role in the design of this study and will not have any role during its execution, analyses, interpretation of the data, or decision to submit results.
Open access funding provided by University of Basel

Data availability
Data are provided within the manuscript or supplementary information files.

Declarations

Ethics approval and consent to participate
The SchistoBreak study protocol was waived by the ethics committee in Switzerland (Ethikkommission Nordwest- und Zentralschweiz; EKNZ) on October 23, 2019 (Req-2019-00951) and approved by the ethics committee in Zanzibar (Zanzibar Health Research Institute; ZAHRI) on December 13, 2019 (ZAHREC/03/PR/December/2019/12). The renewed approvals for the protocol were granted by ZAHRI on December 2, 2020 (ZAHREC/01/RN/December/2020/10) and February 23, 2021 (ZAHREC/04/AMEND/FEB/2021/01). The study was prospectively registered at ISRCTN (ISCRCTN91431493). At the onset of the study and repeatedly before each survey and the intervention period, meetings were held with the community leaders and school principals of the study area to explain research aims and study and intervention procedures. Community leaders and school principals were requested to inform their communities and parents of the schoolchildren about the forthcoming schistosomiasis activities. All individuals participating in the surveys from November 2020 to March 2021 and from November 2021 to March 2022, respectively, and in the 5T intervention period from May 2021 to October 2021 were informed in detail about the objectives and procedures of the study by the local research team and a leaflet that was provided together with informed consent forms. Individuals that were invited to participate in the study could ask questions to the study team. Moreover, the information leaflet included the telephone number of the local principal investigator so that participants or parents of participating children could call in case they had additional questions. Study participants provided written informed consent for their participation. For participating children aged < 18 years, written consent was obtained from their parents or legal guardians. Children aged 12–17 years additionally signed an assent form themselves. Individuals that tested microhaematuria-positive or *S. haematobium*-positive during the annual surveys or 5T activities were offered treatment with a single dose of praziquantel (40 mg/kg body weight).

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

Author details
[1]Swiss Tropical and Public Health Institute, Allschwil, Switzerland
[2]University of Basel, Basel, Switzerland
[3]Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania
[4]Neglected Diseases Program, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania
[5]Neglected Diseases Program, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

Received: 29 November 2023 / Accepted: 21 June 2024
Published online: 02 July 2024

References

1. World Health Organization. (2020). Ending the neglect to attain the sustainable development goals: a road map for neglected tropical diseases 2021–2030. World Health Organization. https://apps.who.int/iris/handle/10665/338565. License: CC BY-NC-SA 3.0 IGO.
2. World Health Organization. Weekly epidemiological record. Schistosomiasis and soil-transmitted helminthiases: progress report. 2021. Geneva: 2022;48:621–32. https://www.who.int/publications/i/item/who-wer9748-621-632
3. World Health Organization. (2022). WHO guideline on control and elimination of human schistosomiasis. https://apps.who.int/iris/handle/10665/351856. License: CC BY-NC-SA 3.0 IGO.
4. Kokaliaris C, Garba A, Matuska M, Bronzan RN, Colley DG, Dorkenoo AM, et al. Effect of preventive chemotherapy with praziquantel on schistosomiasis among school-aged children in sub-saharan Africa: a spatiotemporal modelling study. Lancet Infect Dis. 2022;22(1):136–49. https://doi.org/10.1016/S1473-3099(21)00090-6
5. Barakat RMR. Epidemiology of schistosomiasis in Egypt: travel through time: review. J Adv Res. 2013;4(5):425–32. https://doi.org/10.1016/j.jare.2012.07.003
6. Balahbib A, Amarir F, Bouhout S, Rhajaoui M, Adlaoui E, Sadak A. Review of the urinary schistosomiasis control in Morocco (1960–2018). Interdiscip Perspect Infect Dis. 2020. https://doi.org/10.1155/2020/3868970. 2020:3868970. Epub 2020/10/31.
7. Haggag AA, Rabiee A, Abd Elaziz KM, Gabrielli AF, Abdelhai R, Hashish A, et al. Elimination of schistosomiasis haematobia as a public health problem in five governorates in Upper Egypt. Acta Trop. 2018;188:9–15. https://doi.org/10.1016/j.actatropica.2018.08.024. PubMed PMID: 30145259.
8. Rollinson D, Knopp S, Levitz S, Stothard JR, Tchuem Tchuenté LA, Garba A et al. Time to set the agenda for schistosomiasis elimination. Acta Trop. 2013;128(2):423–40. Epub 2012/05/15. https://doi.org/10.1016/j.actatropica.2012.04.013. PubMed PMID: 22580511.
9. Tchuem Tchuenté LA, Rollinson D, Stothard JR, Molyneux D. Moving from control to elimination of schistosomiasis in sub-saharan Africa: time to change and adapt strategies. Infect Dis Poverty. 2017;6(1):42. https://doi.org/10.1186/s40249-017-0256-8
10. Kouadio JN, Giovanoli Evack J, Sékré JK, Achi LY, Ouattara M, Hattendorf J, et al. Prevalence and risk factors of schistosomiasis and hookworm infection in seasonal transmission settings in northern Côte d'Ivoire: a cross-sectional study. PLoS Negl Trop Dis. 2023;17(7):e0011487. https://doi.org/10.1371/journal.pntd.0011487. Epub 2023/07/17.
11. Trippler L, Knopp S, Welsche S, Webster BL, Stothard JR, Blair L, et al. The long road to schistosomiasis elimination in Zanzibar: a systematic review covering 100 years of research, interventions and control milestones. Adv Parasitol: Academic; 2023;122:71–191. https://doi.org/10.1016/bs.apar.2023.06.001
12. Ramzy RMR, Rabiee A, Abd Elaziz KM, Campbell CH, Kittur N, Colley DG, et al. Test, treat, track, test, and treat active surveillance toward elimination of schistosomiasis: a feasibility study. Am J Trop Med Hyg. 2020;103(4):1572–7. https://doi.org/10.4269/ajtmh.20-0156. Epub 2020/07/15.
13. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-year intervention study on elimination of urogenital schistosomiasis in Zanzibar: parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019;13(5). https://doi.org/10.1371/journal.pntd.0007268
14. Knopp S, Person B, Ame SM, Ali SM, Hattendorf J, Juma S, et al. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: a cluster-randomised trial. Lancet Global Health. 2019;7(8):e1118–29. https://doi.org/10.1016/S2214-109X(19)30189-5
15. Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. Impact of seven years of mass drug administration and recrudescence of *Schistosoma*

haematobium infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies. PLoS Negl Trop Dis. 2021;15(2):e0009127. https://doi.org/10.1371/journal.pntd.0009127. Epub 2021/02/13.
16. Knopp S, Mohammed KA, Ali SM, Khamis IS, Ame SM, Albonico M, et al. Study and implementation of urogenital schistosomiasis elimination in Zanzibar (Unguja and Pemba islands) using an integrated multidisciplinary approach. BMC Public Health. 2012;12:930. https://doi.org/10.1186/1471-2458-12-930
17. Trippler L, Hattendorf J, Ali SM, Ame SM, Juma S, Kabole F, et al. Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study. BMC Infect Dis. 2021;21(1):1024. https://doi.org/10.1186/s12879-021-06620-8. PubMed PMID: 34592960; PubMed Central PMCID: PMCPMC8482678. Epub 2021/10/02.
18. Trippler L, Ali SM, Ame SM, Hattendorf J, Suleiman KR, Ali MN, et al. Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: Baseline cross-sectional survey findings before the onset of a 3-year intervention study. Parasit Vectors. 2022;15(1):292. https://doi.org/10.1186/s13071-022-05404-6. PubMed PMID: 35974353; PubMed Central PMCID: PMCPMC9380971. Epub 2022/08/17.
19. Ministry of Finance and Planning National Bureau of Statistics Tanzania and Presidents' Office - Finance and Planning Office of the Chief Government Statistician Zanzibar. (2022). Administrative Units Population Distribution Report. www.nbs.go.tz/index.php/en/
20. Wizara ya Fedha na Mipango Ofisi ya Taifa ya Takwimu Tanzania. Ofisi ya Rais Fedha na Mipango Ofisi ya Mtakwimu Mkuu wa Serikali Zanzibar. Sensa ya watu na makazi ya mwaka 2022 - matokeo ya mwanzo. 2022.
21. U.S. Food and Drug Administration. Biltricide® tablets (praziquantel) 2010 [26.07.2023]. https://www.accessdata.fda.gov/drugsatfda_docs/label/2010/018714s012lbl.pdf
22. Trippler L, Ali MN, Ame SM, Ali SM, Kabole F, Hattendorf J, et al. GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation. Infect Dis Poverty. 2022;11(1):8. https://doi.org/10.1186/s40249-021-00928-y. PubMed PMID: 35033202; PubMed Central PMCID: PMCPMC8761264. Epub 2022/01/17.
23. Montresor A, Engels D, Ramsan M, Foum A, Savioli L. Field test of the 'dose Pole' for praziquantel in Zanzibar. Trans R Soc Trop Med Hyg. 2002;96(3):323–4. https://doi.org/10.1016/s0035-9203(02)90111-2. Epub 2002/08/15.
24. Savioli L, Albonico M, Amour KA, Saleh JM. Expired reagent strips for haematuria. Trop Doct. 1993;23(4):187–8. https://doi.org/10.1177/004947559302300431. PubMed PMID: 8273181.
25. Grolimund CM, Bärenbold O, Hatz CF, Vennervald BJ, Mayombana C, Mshinda H, et al. Infection intensity-dependent accuracy of reagent strip for the diagnosis of *Schistosoma haematobium* and estimation of treatment prevalence thresholds. PLoS Negl Trop Dis. 2022;16(4):e0010332. https://doi.org/10.1371/journal.pntd.0010332
26. Lo NC, Bezerra FSM, Colley DG, Fleming FM, Homeida M, Kabatereine N, et al. Review of 2022 WHO guidelines on the control and elimination of schistosomiasis. Lancet Infect Dis. 2022;22(11):e327–35. https://doi.org/10.1016/S1473-3099(22)00221-3
27. Maganga JK, Campbell CH, Angelo T, Mosha J, Mwanga JR, Kinung'hi SM. Test-treat-track-test-treat strategy for control of schistosomiasis in two low-prevalence villages in northwestern Tanzania. Am J Trop Med Hyg. 2023;108(6):1167–74. https://doi.org/10.4269/ajtmh.22-0442. Epub 2023/05/10.
28. Celone M, Person B, Ali SM, Lyimo JH, Mohammed UA, Khamis AN, et al. Increasing the reach: involving local muslim religious teachers in a behavioral intervention to eliminate urogenital schistosomiasis in Zanzibar. Acta Trop. 2016;163:142–8. https://doi.org/10.1016/j.actatropica.2016.08.004. PubMed PMID: 27498244; PubMed Central PMCID: PMCPMC5019290.
29. Ndum NC, Trippler L, Mohammed UA, Ali AS, Hattendorf J, et al. Capacities and needs of health care facilities for schistosomiasis diagnosis and management in elimination settings. Parasit Vectors. 2024;17:263. https://doi.org/10.1186/s13071-024-06311-8
30. van der Horst T, Al-Mafazy AW, Fakih BS, Stuck L, Ali A, Yukich J, et al. Operational coverage and timeliness of reactive case detection for malaria elimination in Zanzibar, Tanzania. Am J Trop Med Hyg. 2020;102(2):298–306. https://doi.org/10.4269/ajtmh.19-0505. Epub 2019/11/27.
31. Deutsch-Feldman M, Hamapumbu H, Lubinda J, Musonda M, Katowa B, Searle KM, et al. Efficiency of a malaria reactive test-and-treat program in southern Zambia: a prospective, observational study. Am J Trop Med Hyg. 2018;98(5):1382–8. https://doi.org/10.4269/ajtmh.17-0865. Epub 2018/03/21.
32. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Correction to: urogenital schistosomiasis elimination in Zanzibar: accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections. Parasit Vectors. 2019;12(1):149. https://doi.org/10.1186/s13071-019-3397-8. PubMed PMID: 30922376; PubMed Central PMCID: PMCPMC6440130. Epub 2019/03/30.
33. Krauth SJ, Greter H, Stete K, Coulibaly JT, Traoré SI, Ngandolo BN, et al. All that is blood is not schistosomiasis: experiences with reagent strip testing for urogenital schistosomiasis with special consideration to very-low prevalence settings. Parasit Vectors. 2015;8:584. https://doi.org/10.1186/s13071-015-1165-y. Epub 2015/11/12.
34. King CH, Bertsch D. Meta-analysis of urine heme dipstick diagnosis of *Schistosoma haematobium* infection, including low-prevalence and previously-treated populations. PLoS Negl Trop Dis. 2013;7(9):e2431. https://doi.org/10.1371/journal.pntd.0002431
35. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Urogenital schistosomiasis elimination in Zanzibar: accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections. Parasit Vectors. 2018;11(1):552. https://doi.org/10.1186/s13071-018-3136-6. PubMed PMID: 30352631; PubMed Central PMCID: PMCPMC6199745. Epub 2018/10/26.
36. Gass K. Time for a diagnostic sea-change: rethinking neglected tropical disease diagnostics to achieve elimination. PLoS Negl Trop Dis. 2021;14(12):e0008933. https://doi.org/10.1371/journal.pntd.0008933
37. Toor J, Coffeng LE, Hamley JID, Fronterre C, Prada JM, Castaño MS, et al. When, who, and how to sample: designing practical surveillance for 7 neglected tropical diseases as we approach elimination. J Infect Dis. 2020;221(Suppl 5):S499–502. https://doi.org/10.1093/infdis/jiaa198. Epub 2020/06/13.

Publisher's Note

4. DISCUSSION

Prevalence of *S. haematobium* on Zanzibar has decreased considerably over the last one hundred years [2]. The islands already achieved the elimination of schistosomiasis as a public health problem in 2017 and have been aiming for the interruption of *S. haematobium* transmission ever since [2, 57]. However, an observed spatial heterogeneity of *S. haematobium* infection down to a small administrative area level has raised the need for the improvement of strategies and intervention approaches that move the islands towards the complete elimination of schistosomiasis while addressing the micro-epidemiology of many low-prevalence and a few hotspot areas. This thesis discusses the results of schistosomiasis research in Zanzibar and its implications for future research and policy on the islands and elsewhere.

4.1. The impact of large-scale interventions on schistosomiasis prevalence

The drivers of the major success of eliminating schistosomiasis as a public health problem in Zanzibar were small-scale local pilot projects that were subsequently scaled up, primarily within the last 15 years. This thesis shows that the primary interventions implemented within the last one hundred years covered three central angles: i) treatment, ii) snail control, and iii) behavior change (see 3.1.).

4.1.1. Treatment interventions

During the ZEST study, large-scale preventive chemotherapy was distributed through praziquantel MDA on Unguja and Pemba in schools and communities annually or bi-annually from 2011 to 2017 and resulted in a *S. haematobium* prevalence reduction in 9-12 year old schoolchildren from 4.1% in 2011 to 1.4% in 2017 [58]. There are many more examples of school-based MDA, such as a study in Kenya which showed a *S. mansoni* prevalence reduction from 44.7% to 14.0% after four years [62]. However, just a few other countries and studies have included adults in the MDA. The inclusion of adults in the MDA has only been recommended since 2022 in areas where the prevalence is ≥10% [7].

The Zanzibar islands are among the few regions in the world where adults have already been treated during MDA in the form of community-wide treatment in the past and present [2]. The ZEST study showed that the prevalence in adults was reduced from 3.3% in 2011 to 1.5% in

2017 in the study arm, where MDA was the only intervention (unpublished ZEST study data). Modeling studies show that MDA in school-age children alone will not lead to interruption of *S. haematobium* and *S. mansoni* transmission in high-risk areas [63, 64]. A study in Kenya further supports the importance of MDA in adults as the *S. mansoni* prevalence in adults decreased from 46.9% to 11.9% after four years of community-wide MDA in a high-prevalence area [65].

Further research from the Zanzibar islands also underlines the importance of continuous treatment in areas with high *S. haematobium* prevalence. In 2019, a 16-month treatment gap in praziquantel MDA occurred on the Zanzibar islands resulting in a prevalence rebound in hotspot areas from 2.8% in 2019 to 9.1% in 2020 in school-aged children, while the prevalence in low-prevalence areas stayed considerably low [59]. Likewise, a transmission model based on data from Kenya predicted a likely rebound of *Schistosoma* prevalence after treatment gaps, especially in areas at higher risk [66]. Considering the fast rebound of *Schistosoma* infection after treatment gaps, regular MDA with praziquantel is needed in schoolchildren and adults in hotspot areas in Zanzibar and elsewhere.

Based on the above results and in accordance with the new WHO guidelines, more countries may consider including adults in treatment programs and conducting community MDA in the future. One challenge, however, is the expense associated with drug distribution, which can be particularly high when targeting communities compared to schools where many children can be found and treated in one place [67, 68]. Therefore, funding needs to be secured for implementing MDA in communities to reach the new target group in endemic countries. The more significant challenge, however, may arise from the limited availability and funding for the drugs themselves. Merck KGgA, the manufacturer of praziquantel, has committed to donating the drugs for school-aged children for an unlimited time period, whereas for adults, funding is only secured for certain regions [35]. Countries that had previously targeted only children and may want to include adults in future MDAs will have an increased demand for praziquantel. To meet the demand, the distribution of praziquantel within endemic regions needs to be redesigned, prioritizing its distribution to areas with high prevalence, where MDA is needed the most. For regions with low prevalence, novel alternative treatment schemes, e.g., drug-saving targeted treatment, need to be considered as an alternative to drug-consuming MDA (see 4.3.). If treatment is adapted locally, the demand for praziquantel may be met globally.

Preschool-aged children are another age group that received increased research attention during the last decade. Indeed, in Zimbabwe, a *S. haematobium* prevalence of 45% in children aged 1-5 years was reported in 2020 [69], and in Kenya, a *S. mansoni* prevalence of 11.8% in children aged 3-7 years was reported for 2018 [70]. Since 2022, the WHO has recommended including pre-school-aged children in MDA programs with a coverage of ≥75% [7]. However, the United States Food and Drug Administration does not recommend the treatment of children <4 years old with praziquantel [31]. As no pediatric praziquantel formulation is yet available [71], children <4 years of age remain untreated. However, the Pediatric Praziquantel Consortium has been developing a formulation suitable for young children (arpraziquantel) since 2012 [72]. In brief, the recent phase 3 study showed excellent results in children aged three months to 6 years, and the Consortium expects access to arpraziquantel in sub-Saharan African countries in 2024 [72]. Treating young children would be an important milestone in expanding the coverage of large-scale preventive chemotherapy in hotspot areas in Zanzibar and elsewhere.

4.1.2. Snail control interventions

As long as there have been attempts to treat schistosomiasis in Zanzibar, there have also been attempts to interrupt the human-snail-human life cycle with snail control [2]. As early as 1928, it was suggested to use chemical snail control or to clear swamps of plants to make water bodies less favorable for the intermediate host snails of schistosomiasis [73]. In 1966, the chemical molluscicide niclosamide was used for the first time in Zanzibar [74], which is still the recommended standard molluscicide for snail control [38].

In the 2010s, niclosamide was used in a large-scale cluster-randomized trial on the Zanzibar islands as part of the ZEST study. In a trial arm where niclosamide was applied and MDA was provided, the prevalence in 9-12 year old children decreased from 7.8% in 2011 to 1.7% in 2017 [58]. This result is in line with a study from 2016 evaluating the impact of biological or chemical snail control on *Schistosoma* prevalence in 83 countries/territories in the 20th Century, which found a 92% prevalence reduction in countries/territories where widespread snail control was implemented compared with a 37% reduction in countries/territories with little or no snail control [75]. The study concluded that other interventions, such as treatment and economic factors, may have also contributed to the reduction in prevalence. However, a study in St. Lucia, where snail control with niclosamide was the only intervention, showed a significant decrease

in *S. mansoni* prevalence from 22% in 1970 to 4.3% in 1974/1975 [76], indicating that the application of molluscicides had a tremendous impact on *Schistosoma* prevalence.

Niclosamide has several side effects, including toxicity to fish, invertebrates, and amphibians [7], which is particularly concerning in areas where fishing is a crucial part of the economy. In these areas, a health-impact assessment should be conducted before implementing snail control with niclosamide [38]. Additionally, snail control with niclosamide is expensive [77] and hence, presents a challenge for low-income countries. Various studies have therefore attempted to find alternatives to niclosamide. As early as 1939, experiments with plant extracts were conducted in Zanzibar, and although some of them were successful, they were never tested in a field study in water bodies [2, 78]. Other studies experimented with plant extracts to assess their effectiveness in snail reduction with promising results, including the additional benefit of income to farmers when the plants are cultivated and sold [79]. However, more research needs to be done to explore their future applications fully [79]. Rather than adding a substance that is toxic for the snails to water bodies, a study in Senegal removed vegetation from water bodies that serve as habitats for the intermediate host snails [80]. The study engaged community members in the removal, who then used the vegetation for livestock feeding or as compost to generate income. The study showed a lower *S. mansoni* prevalence in schoolchildren in sites where vegetation was removed than in sites where the vegetation was not removed. The study is an excellent example of capacity development as the community members were encouraged to improve their health while generating income. For Zanzibar and other countries, biological or environmental snail control through the cultivation of plants that are toxic to snails or through the removal of vegetation that serves as snail habitats could be an alternative to the application of niclosamide. However, for each setting, field studies are required first to test the toxicity of native plants and their extracts in the water bodies against snails and other animals and/or to determine which vegetation to remove to yield the most effective snail reduction results.

4.1.3. Behavior change interventions

Interventions targeting behavior change in individuals on the Zanzibar islands have transitioned from a top-down health education approach in the early 20th Century to a behavioral communication approach in the 21st Century, in which the community has been involved in needs assessments and focus group discussions [2, 81]. The latter underpinned the ZEST study

and was followed by large-scale behavioral interventions in 30 areas of Unguja and Pemba, including the construction of washing platforms and latrines and education in schools and communities [58, 82]. Due to low overall prevalence on the islands, the stand-alone impact of behavioral interventions in the ZEST study was never significantly shown but has likely contributed to the decrease of prevalence in school-aged children from 6.4% in 2011 to 1.6% in 2017 in the areas where the interventions were implemented together with MDA [58]. This is consistent with a review concluding that behavior change interventions, when adapted to the local settings and conditions, may support reducing *Schistosoma* prevalence [25].

Behavior change interventions in Zanzibar have been traditionally conducted in schools and within centers of communities [2]. A study in Nigeria identified additional possible outreach grounds, including mechanic shops and football grounds [83]. Football and other sports are an important part of people's lives in Zanzibar, although predominantly in men (own observations). However, since prevalence of schistosomiasis has almost always been higher in men/boys than in women/girls in Zanzibar [2], football grounds could be a future site for targeted behavior change interventions. This thesis also shows that the prevalence of microhematuria as a proxy for *S. haematobium* infection in Zanzibar is particularly high among patients in health facilities (see 3.6.). Hence, implementing behavior change interventions in health facilities could be another essential place to reach at-risk people. Here, primarily women could be reached, especially when targeting health facilities that include a mother and child care clinic. Preventing *Schistosoma* infection in pregnant women is crucial, as studies have shown that schistosomiasis may contribute to anemia, particularly in cases of heavy infection [84, 85]. Anemia in pregnant women is associated with low birth weight and maternal death [86, 87]. In addition, a study in Kenya showed that two-year-old children born to *S. mansoni*-infected mothers had a significantly lower response to measles vaccination than children born to not infected mothers [88]. Including additional intervention sites in behavior change interventions, such as football grounds or health facilities, might be vital for future interventions.

Community engagement and the involvement of community and religious leaders are essential for a change of behavior in the population. Therefore, focus group discussions and needs assessments were conducted to inform the behavior change interventions within the ZEST study and were also the basis for the behavior change interventions in the SchistoBreak study [58, 60, 89]. Much was learned from these assessments; however, they were conducted 12 years ago, and it is time for a new evaluation of the perceptions, needs, and wishes of communities

in Zanzibar to prevent *Schistosoma* infections. Nonetheless, sustainable behavior change can only be achieved if the infrastructure is in place to provide a good alternative for using safe water, such as for washing clothes or bathing. A study in Namibia showed that the risk of contracting schistosomiasis or soil-transmitted helminthiasis is significantly 1.5-fold higher in individuals without access to potable water and doubled if there is no access to sanitation infrastructure [90]. Significant investments in infrastructure by the government or donors are needed to enable real behavior change. In addition, the community could be involved in the time and financial investments for new infrastructure. A study in Uganda assessed the willingness of community members to invest money and time in the construction of WASH resources, such as clean water taps in the village [91]. The study indicated that the level of community engagement and willingness to contribute time to the construction of WASH infrastructure is higher when the knowledge about water-borne diseases, such as schistosomiasis, is high. The results of the study conducted in Uganda demonstrate the significance of implementing behavior change interventions in effectively transferring knowledge about the *Schistosoma* life cycle and how the interventions can strengthen community involvement and capacity development. In Zanzibar, continuous behavior change interventions are essential, especially in hotspot areas, and it is time for a new assessment of the perception and needs regarding schistosomiasis interventions. In addition, the evaluation could include questions about the community members' willingness to contribute to constructing new WASH infrastructure.

A combination of treatment, snail control, and behavior change is crucial for hotspot areas to reduce prevalence further, however all three interventions could be optimized for implementation for Zanzibar and other countries: i) once funding and drug procurement is secured, it is essential to also include adults and preschool-aged children in MDA through community-wide treatment. ii) new snail control measures need to be identified that are more environmentally friendly than niclosamide but equally effective, iii) regular needs assessments and inclusion of community members in decision-making processes need to be embedded in behavior change measures, for which new intervention grounds may be included to reach many people.

4.2. Micro-mapping of infection and environmental risk factors

There has long been a paucity of georeferenced infection data for schistosomiasis [37, 92], but in recent decades, more studies have georeferenced their infection data, including the SchistoBreak study. In the SchistoBreak study, *S. haematobium* infection is assessed at the school and household level, and these sites are georeferenced, allowing to identify small-scale infection hotspots [93, 94]. All grades are included in schools, and an equal number of children is randomly selected per grade; in households, the entire household is invited to participate. Many other approaches to spatial sampling exist. Another study that focused on micro-mapping of *S. mansoni* and *S. haematobium* infection took place in Kenya and used a different approach by selecting villages and randomly selecting children aged 8-14 years in randomly selected households [95]. The selection of villages was risk-based as proximity to water bodies was a factor, and villages closer to water bodies were selected first. A study in Ethiopia chose to micro-map *S. mansoni* and *S. haematobium* infections by randomly selecting households based on so-called family folders at the post office and selecting one household member from one of five predefined age groups by sex [96]. These three studies illustrate the variety of parameters used to select study participants and different age groups for mapping *Schistosoma* infections. While there is much to be learned from the different methods, it is challenging to compare prevalence across regions if the approaches to assessing prevalence are different.

Micro-mapping may be adapted to the local context and resources, e.g., while the remote roads in Pemba do not have names, other regions/countries may have street names based on which random household selection may be made. However, WHO guidance on how to subdivide age groups in children and adults and whether it is best to survey schools and/or whole households or selected individuals from households may help to make prevalence estimates more consistent and comparable across regions or countries. Data collected in a consistent manner enhances the global assessment of infected individuals and supports a framework for monitoring and evaluating progress that is needed to validate whether geographical areas have actually achieved the WHO goals of “elimination of schistosomiasis as a public health problem” or “interruption of *Schistosoma* transmission” [97]. Currently, the WHO only reports the number of people requiring preventive chemotherapy, and there is no up-to-date estimate of infections worldwide [28]. The lack of reported global infection numbers for *Schistosoma* may be because there has been no systematic analysis of the worldwide distribution of *Schistosoma* infection since 2006 [50]. An infection estimate for sub-Saharan Africa was

created in 2022, but a global assessment is still lacking [26]. Since all species of *Schistosoma*, including those primarily found in Asia and the Americas, are treated with praziquantel [1], an updated estimate of the number of people infected worldwide is urgently needed to enable policymakers to make informed decisions on, for example, the global distribution of praziquantel (see 4.1.1).

Micro-mapping can be enhanced not only by georeferencing human infection data but also by assessing environmental factors that may impact the transmission of *Schistosoma* or other NTDs. As part of the SchistoBreak study, the locations of freshwater bodies (with and without detected *B. globosus*) in northern Pemba were georeferenced and provided invaluable information about possible transmission sites [93]. A study in 2016 assessed the association of environmental factors with the *B. globosus* abundance on Unguja but only found the presence of grass at water bodies as a significant factor for *B. globosus* abundance [98]. However, the study did not assess snail abundance over time and seasons or consider broader environmental factors such as rainfall or seasonal temperature. A study conducted in South Africa found that rainfall, pH, habitat type, abundance of other freshwater snails, and seasons influenced the distribution, abundance, and/or infection rates of *B. globosus* [99]. However, due to the specific habitat preferences of different *Bulinus* species across sub-Saharan Africa and the resulting genetic divergence [100], it is still unknown whether the results of studies conducted elsewhere can simply be applied to the Zanzibar islands. There remains a need to understand how environmental factors predict snail density and, thus, the risk of transmission. Since regular snail surveys have been conducted and environmental factors have been assessed within the SchistoBreak study, analyses of these data, integrated with external seasonal data and rainfall patterns, may yield results that indicate peak *Schistosoma* transmission seasons on the Zanzibar islands.

Another approach to model environmental data was used in a study to micro-map onchocerciasis and loiasis in Central and West Africa, using geographical information system software to examine high-resolution satellite imagery [101]. The study identified low and high-risk infection transmission areas by layering environmental and prevalence data [101]. This method dramatically reduced cost and time compared to field surveys, especially in remote and inaccessible areas where field surveys would be difficult to conduct. A study in Zambia that aimed to map water bodies for potential malaria vector breeding sites took remote sensing micro-mapping to a new level by adapting the mapping to the seasons and even predicting vegetated water bodies from satellite data [102]. The study was able to map open and vegetated

water bodies with up to 97% accuracy. Remote sensing and geographical information systems have existed since the 1970s, but high-resolution satellite imagery is a recent development [103, 104]. Strategies such as those employed in the studies in Cameroon and Zambia can potentially be used for micro-mapping and transmission risk assessment of any disease where the occurrence of the vector is linked to environmental factors, such as water bodies for schistosomiasis or onchocerciasis, as long as access to high-resolution imagery and analysis expertise are available.

Considerable progress has been made in micro-mapping the prevalence of schistosomiasis and other NTDs, and new tools and technologies even allow the prediction of hotspot and low-prevalence areas. While much can be learned from different mapping approaches for schistosomiasis, NTDs, and other diseases, guidance by the WHO on how to collect data in a consistent manner may help to make prevalence estimates more consistent and comparable across regions or countries. The guidelines could be embedded in a framework for monitoring and evaluating progress to validate whether geographical areas have actually achieved the WHO goals of "elimination of schistosomiasis as a public health problem" or "interruption of transmission" [97]. Furthermore, the creation of a new *Schistosoma* infection world map would be promoted, which is urgently needed to adjust interventions locally so there are enough resources globally. Finally, micro-mapping enables the implementation of interventions targeted to the local micro-epidemiology.

4.3. Targeted alternatives to large-scale interventions in low-prevalence settings

The very low *S. haematobium* prevalence in Zanzibar has raised the question of overtreatment through MDA in a mostly healthy population [2]. The SchistoBreak study introduced mapping of *S. haematobium* infections as a strategy to implement interventions risk-based and focally [93, 94]. The proximity of schools and households to freshwater bodies with *B. globosus* has been found to have a tremendous impact on the chance of schoolchildren and community members being infected [93]. Due to the mapping, an alternative approach to MDA could be implemented in low-prevalence areas in the north of Pemba in 2021: test-treat-track-test-treat (5T) interventions with risk-based testing of children in primary and Islamic schools in close proximity to water bodies [60]. Since the study is ongoing, no final recommendation can be made about the feasibility of the interventions to keep prevalence low in the long term or even advance towards interruption of transmission. Nonetheless, tracking positive index cases identified many additional infected individuals, and after one year of 5T interventions instead of MDA, the *S. haematobium* prevalence did not increase significantly despite no MDA. These results show a promising outlook for the future, and if the final results of the SchistoBreak study support the positive results, the same or similarly adapted interventions could be implemented in other regions in Zanzibar and elsewhere where prevalence of schistosomiasis, other NTDs, or diseases like malaria, is very low.

While there are only a few other instances of 5T interventions serving as an alternative to MDA in the field of schistosomiasis (see 3.6.), studies on other infectious diseases provide examples of alternative interventions. One example of alternative intervention implementation comes from a study conducted in mainland Tanzania that focused on taeniosis [105]. For the study, collection points in villages were set up, and individuals were invited to provide a stool sample for testing for taeniosis. Whoever tested positive for taeniosis was subsequently treated. Although the aim of the study was not to identify an alternative to MDA, it presents an example of a test-and-treat approach that could be potentially implemented for schistosomiasis test-and-treat in Zanzibar and elsewhere. Various collection points set up for one day each in low-prevalence areas provide an easily accessible possibility for community members to get tested at the point-of-care. Participants of the SchistoBreak study indicated concerns about the lack of medical personnel among the individuals delivering the drugs during the MDA [93]. To avoid testing hesitancy and to strengthen the collaboration with health facilities, a future study could investigate whether individuals are willing to get tested at these collection points,

especially when medical personnel are present. To attract many participants, this approach could be combined with a "schistosomiasis day" for the whole community based on the example of "kichocho-days" conducted in schools in the ZEST study and in the hotspot implementation units of the SchistoBreak study [60, 89]. Such an event would provide an opportunity for behavior change intervention and motivate individuals to provide their urine samples. Involving religious and village leaders may further enhance the motivation in the community [106].

Another alternative to large-scale MDA comes from a study on malaria in Eswatini. In the study, individuals who tested positive at health facilities were tracked to their households and focal MDA was conducted by treating the whole household and surrounding households for malaria without prior testing [107]. Such an approach could be embedded in the health care system and into schistosomiasis interventions in Zanzibar and elsewhere. The testing material and drug procurement would need to be secured for all health facilities while ensuring correspondence between the health facilities and teams that track household members. However, in Zanzibar, this approach might be challenging due to the growing treating-without-testing hesitancy in the population [93], making it difficult to treat the household members and surrounding households without testing beforehand.

Focal MDA or focal test-and-treat interventions could also be based on geographical factors instead of identified index cases. The mapping within the SchistoBreak study revealed that the chance of getting infected is highest for those living close (<1 km) to water bodies where *B. globosus* was found. Hence, a viable alternative to MDA could be the treatment of all individuals living within <1 km distance to the water body where the presence of *B. globosus* is confirmed. This approach would require continuous malacological surveys since the presence of *B. globosus* cannot (yet) be predicted based on environmental factors (see 4.2.). However, in comparison to large-scale MDA, this approach would be drug-saving. Another less-drug-saving approach, but still more drug-saving than large-scale MDA, could be to use remote sensing and satellite imagery to map all water bodies (without confidently knowing whether or not the intermediate host snails are present) and treat all individuals living within <1 km distance to any water body. Given the expenses for continuous island-wide malacological surveys, this approach may be a viable alternative. Once a prediction of snail habitats using collected data and remote sensing is possible for Zanzibar (see 4.2.), the treatment could be concentrated on the 1 km radius around the water bodies with the highest likelihood of being an intermediate host snail habitat. To find the households within a 1 km

radius around the water bodies for conducting the focal MDA or focal test-and-treat interventions, a navigation tablet-based tool like the one used to identify the pre-randomized houses for the community-based household survey in the SchistoBreak study [94], could be used. All approaches that involve testing first could be considerably more efficient if a highly sensitive and specific diagnostic test would be available that could be used at the point-of-care.

Interventions cannot only be targeted to the infected individuals but also to the intermediate host snail. Malacological surveys at water bodies suspected of being habitats of intermediate host snails can identify the water bodies as transmission sites. When intermediate host snail presence is confirmed, focal snail control with niclosamide, as opposed to its indiscriminate application, minimizes the impact on the ecosystem [2]. A study conducted in St. Lucia supports the focal application of niclosamide as it concludes that niclosamide is most effective if applied highly focally [76]. Furthermore, when applied in conjunction with the peak transmission season, the effect of molluscicide can be maximized while minimizing frequency and adverse effects [108]. Currently, snail control is focally applied on Pemba as part of the SchistoBreak study [60], but is not linked to the peak transmission season due to the scarcity of evidence on the environmental factors that affect snail abundance in Zanzibar (see 4.2.). Once the evidence is provided, this knowledge can guide niclosamide application to be timed to the peak *Schistosoma* transmission. Moreover, based on studies showing that the ideal time for MDA would be during the low-transmission season, near the minimum snail density [109, 110], MDA and targeted treatment interventions could be timed to seasonal transmission and snail control. Thus, by gaining broader in-depth knowledge of local transmission patterns, the timing of future interventions in Zanzibar, such as treatment and snail control, could be optimized.

Questions have been raised about whether, and at what prevalence level, MDA becomes unethical and too expensive. Furthermore, continuing large-scale treatment in areas with low *Schistosoma* prevalence results in overtreatment of a mostly healthy population. Alternative interventions need to be explored that can be implemented in these areas and maintain the low prevalence. Alternatives can include focal treatment and focal snail control, implemented in locations with the highest risk of transmission to save resources. If focal treatment is paired with testing, the testing opportunity needs to be easily accessible to the population. Furthermore, test-and-treat interventions may be conducted in conjunction with medical personnel of health facilities to lower hesitance to participate and to strengthen collaboration with local health facilities. Focal snail control would be improved with knowledge about the

transmission peak and the variables influencing the abundance of intermediate host snails in water bodies. Therefore, focal treatment and focal snail control interventions can be effectively implemented in conjunction when focusing on the same locations with high transmission risk.

5. CONCLUSION

In the long history of schistosomiasis research and interventions on the Zanzibar islands, the primarily implemented interventions included treatment, snail control, and measures to change behavior, and research showed that future policy for hotspot areas needs to include a combination of all three interventions. Additionally, new WASH infrastructure that serves as an alternative to using freshwater bodies needs to be realized. While the Zanzibar islands already include the treatment of adults in the community-wide MDA, including adults and preschool-aged children is also crucial for hotspot areas in other countries once funding and drug procurement is secured. Snail control with niclosamide is an important intervention to reduce *Schistosoma* prevalence, but future research needs to identify new snail control measures that are more environmentally friendly. Behavior change interventions have proven to be important for future multi-disciplinary interventions in sharing knowledge about the life cycle and prevention measures. Here, new intervention grounds, such as football courts or health facilities, could be crucial to reach more people. Furthermore, an updated needs assessment in the community is vital to enable the community to participate in decision-making and intervention realization, such as the building and maintenance of new infrastructure.

Considerable progress has been made in the strategies to micro-map the prevalence of schistosomiasis and other NTDs, and new tools and technologies even enable the prediction of hotspot and low-prevalence areas. Many studies have used different approaches for prevalence assessment, including diverse selections of study participants, but standard guidelines by the WHO on how to micro-map prevalence for NTDs is now needed to enable the comparison of prevalence across different countries and regions. These standard guidelines embedded in a framework for monitoring and evaluating progress would enhance the validation of whether geographical areas have actually achieved the WHO goals of "elimination of schistosomiasis as a public health problem" or "interruption of transmission". Furthermore, an updated global map of schistosomiasis and/or NTD prevalence would allow an optimal global resource allocation. Finally, micro-mapping enables the implementation of interventions targeted to the

local micro-epidemiology of schistosomiasis and, if transmission patterns are known, targeted to the seasonal transmission of *Schistosoma* spp.

Targeted interventions as an alternative to MDA for low-prevalence areas are required to avoid overtreatment of a primarily healthy population and to set resources free for regions where they are needed most, for countries and regions with high schistosomiasis prevalence. To optimize a resource allocation globally, interventions need to be tailored to the epidemiology locally. The 5T interventions of the SchistoBreak study are an excellent example of a novel approach of targeted interventions, including focal testing, treating, and focal snail control in low-prevalence areas. Results of future study years of the SchistoBreak study and other studies will reveal the optimal targeted interventions needed in various elimination settings to maintain low prevalence levels and to further reduce prevalence towards the goal of interruption of transmission.

Main policy recommendations

- Combination of MDA (for preschool-aged children, school-aged children and adults), snail control and behavior change interventions in hotspot areas
- Governmental investments in water, sanitation and hygiene infrastructure in schistosomiasis-endemic settings
- Creation of WHO guidelines for assessing prevalence of schistosomiasis and other NTDs in a consistent manner

Main research needs

- Identification of highly effective snail control that is environmentally friendly
- Exploration of new intervention grounds for behavior change that fit local settings
- Assessment of seasonal transmission patterns of *S. haematobium* in Zanzibar
- Determination of whether 5T interventions maintain prevalence levels in low-prevalence areas in Zanzibar in the long term and ultimately whether they support reaching the goal of interruption of *S. haematobium* transmission

Research at the Swiss TPH follows the value chain of innovation-validation-application and this thesis has demonstrated that schistosomiasis and snail-related research on the Zanzibar islands often already progressed from innovation to validation and finally to the application of interventions in Zanzibar and elsewhere. Several times innovative research conducted on the

islands has shaped the policy for intervention implementation on the islands and elsewhere and has highly contributed to the islands now aiming to interrupt *S. haematobium* transmission. With the SchistoBreak study and interruption of *S. haematobium* transmission in sight, a new era of innovation and improvement of strategies and intervention approaches for schistosomiasis elimination has started in Zanzibar. Multi-disciplinary interventions in hotspot areas, the novel and innovative micro-mapping strategies, and the targeted interventions in low-prevalence areas within the SchistoBreak study may guide the road down to zero *S. haematobium* infections. First results of the study already validated the positive impact of the interventions on schistosomiasis prevalence. Future study years will show whether the interventions are feasible and impactful in the long term and whether the SchistoBreak study presents another example of Zanzibar research where innovation has led to application with policy recommendations across Zanzibar and outside the island's borders.

REFERENCES

1. Colley DG, Bustinduy AL, Secor WE, King CH. Human schistosomiasis. Lancet. 2014;383(9936):2253-64. doi: https://doi.org/10.1016/S0140-6736(13)61949-2.

2. Trippler L, Knopp S, Welsche S, Webster BL, Stothard JR, Blair L, et al. The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones. Adv Parasitol: Academic Press; 2023. doi: https://doi.org/10.1016/bs.apar.2023.06.001.

3. Colley DG, Secor WE. Immunology of human schistosomiasis. Parasite Immunol. 2014;36(8):347-57. doi: https://doi.org/10.1111/pim.12087.

4. Gryseels B, Polman K, Clerinx J, Kestens L. Human schistosomiasis. Lancet. 2006;368(9541):1106-18. doi: https://doi.org/10.1016/s0140-6736(06)69440-3.

5. Brown DS. Freshwater snails of Africa and their medical importance (1st ed.). CRC Press. 1994. doi: https://doi.org/10.1201/9781482295184.

6. Nation CS, Da'dara AA, Marchant JK, Skelly PJ. Schistosome migration in the definitive host. PLoS Negl Trop Dis. 2020;14(4):e0007951. doi: https://doi.org/10.1371/journal.pntd.0007951.

7. World Health Organization. (2022). WHO guideline on control and elimination of human schistosomiasis. Available from: https://apps.who.int/iris/handle/10665/351856. License: CC BY-NC-SA 3.0 IGO

8. World Health Organization. Prevention and control of schistosomiasis and soil-transmitted helminthiasis: report of a WHO expert committee. World Health Organization, 2002. Available from: https://apps.who.int/iris/bitstream/handle/10665/42588/WHO_TRS_912.pdf?sequence=1&isAllowed=y

9. Knopp S, Ame SM, Hattendorf J, Ali SM, Khamis IS, Bakar F, et al. Correction to: Urogenital schistosomiasis elimination in Zanzibar: Accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections (Parasites & Vectors (2018) 11 (552) DOI: 10.1186/s13071-018-3136-6). Parasit Vectors. 2019;12(1). doi: https://doi.org/10.1186/s13071-019-3397-8.

10. Bogoch, II, Koydemir HC, Tseng D, Ephraim RKD, Duah E, Tee J, et al. Evaluation of a mobile phone-based microscope for screening of *Schistosoma haematobium* infection in rural Ghana. Am J Trop Med Hyg. 2017;96(6):1468-71. doi: https://doi.org/10.4269/ajtmh.16-0912.

11. Krauth SJ, Greter H, Stete K, Coulibaly JT, Traoré SI, Ngandolo BN, et al. All that is blood is not schistosomiasis: Experiences with reagent strip testing for urogenital schistosomiasis with special consideration to very-low prevalence settings. Parasit Vectors. 2015;8:584. doi: https://doi.org/10.1186/s13071-015-1165-y.

12. Bassiouny HK, Hasab AA, El Nimr NA, Al Shibani LA, Al Waleedi AA. (2014). Rapid diagnosis of schistosomiasis in Yemen using a simple questionnaire and urine reagent strips. World Health Organization, Regional Office for the Eastern Mediterranean. Available from: https://apps.who.int/iris/handle/10665/204150.

13. Mazigo HD, Mwingira UJ, Zinga MM, Uisso C, Kazyoba PE, Kinung'hi SM, et al. Urogenital schistosomiasis among pre-school and school aged children in four districts of north western Tanzania after 15 years of mass drug administration: Geographical prevalence, risk factors and performance of haematuria reagent strips. PLoS Negl Trop Dis. 2022;16(10):e0010834. doi: https://doi.org/10.1371/journal.pntd.0010834.

14. Grolimund CM, Bärenbold O, Hatz CF, Vennervald BJ, Mayombana C, Mshinda H, et al. Infection intensity-dependent accuracy of reagent strip for the diagnosis of *Schistosoma haematobium* and estimation of treatment prevalence thresholds. PLoS Negl Trop Dis. 2022;16(4):e0010332. doi: https://doi.org/10.1371/journal.pntd.0010332.

15. King CH, Bertsch D. Meta-analysis of urine heme dipstick diagnosis of *Schistosoma haematobium* infection, including low-prevalence and previously-treated populations. PLoS Negl Trop Dis. 2013;7(9):e2431. doi: https://doi.org/10.1371/journal.pntd.0002431.

16. Ochodo EA, Gopalakrishna G, Spek B, Reitsma JB, van Lieshout L, Polman K, et al. Circulating antigen tests and urine reagent strips for diagnosis of active schistosomiasis in endemic areas. Cochrane Database Syst Rev. 2015;2015(3):Cd009579. doi: https://doi.org/10.1002/14651858.CD009579.pub2.

17. Delgado MK, Delgado EM. 113 - Hematuria. In: Adams JG, editor. Emergency Medicine (Second Edition). Philadelphia: W.B. Saunders; 2013. p. 984-9.e1. doi: https://doi.org/10.1016/B978-1-4377-3548-2.00113-0.

18. Corstjens PLAM, de Dood CJ, Knopp S, Clements MN, Ortu G, Umulisa I, et al. Circulating anodic antigen (CAA): A highly sensitive diagnostic biomarker to detect active *Schistosoma* infections—improvement and use during SCORE. American Journal of Tropical Medicine and Hygiene. 2020;103:50-7. doi: https://doi.org/10.4269/ajtmh.19-0819.

19. Rostron P, Pennance T, Bakar F, Rollinson D, Knopp S, Allan F, et al. Development of a recombinase polymerase amplification (RPA) fluorescence assay for the detection of *Schistosoma haematobium*. Parasit Vectors. 2019;12(1):514-. doi: https://doi.org/10.1186/s13071-019-3755-6.

20. Guegan H, Fillaux J, Charpentier E, Robert-Gangneux F, Chauvin P, Guemas E, et al. Real-time PCR for diagnosis of imported schistosomiasis. PLoS Negl Trop Dis. 2019;13(9):e0007711. doi: https://doi.org/10.1371/journal.pntd.0007711.

21. Turner P, Lalloo K, Bligh J, Armstrong M, Whitty CJ, Doenhoff MJ, et al. Serological speciation of human schistosome infections by ELISA with a panel of three antigens. J Clin Pathol. 2004;57(11):1193-6. doi: https://doi.org/10.1136/jcp.2003.014779.

22. Cherkaoui D, Mesquita SG, Huang D, Lugli EB, Webster BL, McKendry RA. CRISPR-assisted test for *Schistosoma haematobium*. Sci Rep. 2023;13(1):4990. doi: https://doi.org/10.1038/s41598-023-31238-y.

23. Molehin AJ, McManus DP, You H. Vaccines for human schistosomiasis: Recent progress, new developments and future prospects. Int J Mol Sci. 2022;23(4). doi: https://doi.org/10.3390/ijms23042255.

24. King CH, Sutherland LJ, Bertsch D. Systematic review and meta-analysis of the impact of chemical-based mollusciciding for control of *Schistosoma mansoni* and *S. haematobium* transmission. PLoS Negl Trop Dis. 2015;9(12):e0004290. doi: https://doi.org/10.1371/journal.pntd.0004290.

25. Torres-Vitolas CA, Trienekens SCM, Zaadnoordijk W, Gouvras AN. Behaviour change interventions for the control and elimination of schistosomiasis: A systematic review of evidence from low- and middle-income countries. PLoS Negl Trop Dis. 2023;17(5):e0011315. doi: https://doi.org/10.1371/journal.pntd.0011315.

26. Kokaliaris C, Garba A, Matuska M, Bronzan RN, Colley DG, Dorkenoo AM, et al. Effect of preventive chemotherapy with praziquantel on schistosomiasis among school-aged

children in sub-Saharan Africa: a spatiotemporal modelling study. The Lancet Infectious Diseases. 2022;22(1):136-49. doi: https://doi.org/doi.org/10.1016/S1473-3099(21)00090-6.

27. Fifty-fourth World Health Assembly WHA54.19. Schistosomiasis and soil-transmitted helminth infections. 2001. Available from: https://apps.who.int/iris/bitstream/handle/10665/78794/ea54r19.pdf

28. World Health Organization. Weekly epidemiological record. Schistosomiasis and soil-transmitted helminthiases: progress report, 2021. Geneva: 2022. 48. 621-32. Available from: https://www.who.int/publications/i/item/who-wer9748-621-632

29. World Health Organization. Preventive chemotherapy in human helminthiasis: Coordinated use of anthelminthic drugs in control interventions: a manual for health professionals and programme managers. Geneva: World Health Organization, 2006. Available from: https://apps.who.int/iris/bitstream/handle/10665/43545/9241547103_eng.pdf?sequence=1&isAllowed=y

30. Montresor A, Engels D, Ramsan M, Foum A, Savioli L. Field test of the 'dose pole' for praziquantel in Zanzibar. Trans R Soc Trop Med Hyg. 2002;96(3):323-4. doi: https://doi.org/10.1016/s0035-9203(02)90111-2.

31. U.S. Food and Drug Administration. Biltricide® tablets (praziquantel) 2010 [26.07.2023]. Available from: https://www.accessdata.fda.gov/drugsatfda_docs/label/2010/018714s012lbl.pdf.

32. Caldwell N, Afshar R, Baragaña B, Bustinduy AL, Caffrey CR, Collins JJ, et al. Perspective on schistosomiasis drug discovery: highlights from a schistosomiasis drug discovery workshop at Wellcome Collection, London, September 2022. ACS Infect Dis. 2023;9(5):1046-55. doi: https://doi.org/10.1021/acsinfecdis.3c00081.

33. Aboagye IF, Addison YAA. Praziquantel efficacy, urinary and intestinal schistosomiasis reinfection – A systematic review. Pathog Glob Health. 2022:1-8. doi: https://doi.org/10.1080/20477724.2022.2145070.

34. World Health Assembly. (2001). Fifty-fourth World Health Assembly, Geneva, 14-22 May 2001: resolutions and decisions. World Health Organization. Available from: https://apps.who.int/iris/handle/10665/260183. http://apps.who.int/iris/

35. Merck provides 1.5 billionth tablet of praziquantel for treatment of schistosomiasis [Internet]. Darmstadt, Germany; 2022. Available from: https://www.merckgroup.com/en/news/praziquantel-tablet-donation-24-01-2022.html

36. World Health Organization. (2020). Ending the neglect to attain the sustainable development goals: a road map for neglected tropical diseases 2021–2030. World Health Organization. Available from: https://apps.who.int/iris/handle/10665/338565. License: CC BY-NC-SA 3.0 IGO

37. Rollinson D, Knopp S, Levitz S, Stothard JR, Tchuem Tchuenté LA, Garba A, et al. Time to set the agenda for schistosomiasis elimination. Acta Trop. 2013;128(2):423-40. doi: https://doi.org/10.1016/j.actatropica.2012.04.013.

38. World Health Organization. Field use of molluscicides in schistosomiasis control programmes - an operational manual for programme managers. 2017. 50. Available from: https://www.who.int/publications/i/item/9789241511995

39. Grimes JET, Croll D, Harrison WE, Utzinger J, Freeman MC, Templeton MR. The relationship between water, sanitation and schistosomiasis: A systematic review and meta-analysis. PLoS Negl Trop Dis. 2014;8(12):e3296. doi: https://doi.org/10.1371/journal.pntd.0003296.

40. Secor WE. Water-based interventions for schistosomiasis control. Pathog Glob Health. 2014;108(5):246-54. doi: https://doi.org/10.1179/2047773214y.0000000149.

41. Anyolitho MK, Poels K, Huyse T, Tumusiime J, Mugabi F, Tolo CU, et al. Knowledge, attitudes, and practices regarding schistosomiasis infection and prevention: A mixed-methods study among endemic communities of western Uganda. PLoS Negl Trop Dis. 2022;16(2):e0010190. doi: https://doi.org/10.1371/journal.pntd.0010190.

42. Burke ML, Jones MK, Gobert GN, Li YS, Ellis MK, McManus DP. Immunopathogenesis of human schistosomiasis. Parasite Immunol. 2009;31(4):163-76. doi: https://doi.org/10.1111/j.1365-3024.2009.01098.x.

43. World Health Organization. 34th meeting of the International Task Force for Disease Eradication, 19-20 September 2022. 2022. Contract No.: WER No 04, 2023, 98. 41–50. Available from: https://www.who.int/publications/i/item/who-wer9804-41-50

44. Efared B, Bako ABA, Idrissa B, Alhousseini D, Boureima HS, Sodé HC, et al. Urinary bladder *Schistosoma haematobium*-related squamous cell carcinoma: a report of two fatal cases

and literature review. Tropical Diseases, Travel Medicine and Vaccines. 2022;8(1):3. doi: https://doi.org/10.1186/s40794-022-00161-x.

45. Bustinduy AL, Randriansolo B, Sturt AS, Kayuni SA, Leutscher PDC, Webster BL, et al. Chapter One - An update on female and male genital schistosomiasis and a call to integrate efforts to escalate diagnosis, treatment and awareness in endemic and non-endemic settings: The time is now. In: Rollinson D, Stothard R, editors. Adv Parasitol. 115: Academic Press; 2022. p. 1-44. doi: https://doi.org/10.1016/bs.apar.2021.12.003.

46. Kjetland EF, Leutscher PDC, Ndhlovu PD. A review of female genital schistosomiasis. Trends Parasitol. 2012;28(2):58-65. doi: https://doi.org/10.1016/j.pt.2011.10.008.

47. Sturt AS, Webb EL, Francis SC, Hayes RJ, Bustinduy AL. Beyond the barrier: Female genital schistosomiasis as a potential risk factor for HIV-1 acquisition. Acta Trop. 2020;209:105524. doi: https://doi.org/10.1016/j.actatropica.2020.105524.

48. Kayuni S, Lampiao F, Makaula P, Juziwelo L, Lacourse EJ, Reinhard-Rupp J, et al. A systematic review with epidemiological update of male genital schistosomiasis (MGS): A call for integrated case management across the health system in sub-Saharan Africa. Parasite Epidemiology and Control. 2019;4:e00077. doi: https://doi.org/10.1016/j.parepi.2018.e00077.

49. World Health Organization. (2023). Global report on neglected tropical diseases. Available from: https://www.who.int/publications/i/item/9789240067295. CC BY-NC-SA 3.0 IGO

50. Steinmann P, Keiser J, Bos R, Tanner M, Utzinger J. Schistosomiasis and water resources development: Systematic review, meta-analysis, and estimates of people at risk. Lancet Infect Dis. 2006;6(7):411-25. doi: https://doi.org/10.1016/s1473-3099(06)70521-7.

51. Wami WM, Nausch N, Midzi N, Gwisai R, Mduluza T, Woolhouse M, et al. Identifying and evaluating field indicators of urogenital schistosomiasis-related morbidity in preschool-aged children. PLoS Negl Trop Dis. 2015;9(3):e0003649. doi: https://doi.org/10.1371/journal.pntd.0003649.

52. Ministry of Finance and Planning - National Bureau of Statistics Tanzania, President's Office - Finance and Planning - Office of the Chief Government Statistician Zanzibar. (2022). Administrative units population distribution report. Available from: www.nbs.go.tz/index.php/en/.

53. Wizara ya Fedha na Mipango Ofisi ya Taifa ya Takwimu Tanzania, Ofisi ya Rais Fedha na Mipango Ofisi ya Mtakwimu Mkuu wa Serikali Zanzibar. Sensa ya watu na makazi ya mwaka 2022 - matokeo ya mwanzo. 2022.

54. Office of Chief Government Statistician Zanzibar. Main report 2019/20 household budget survey. 2020. Available from: https://www.ocgs.go.tz

55. NOAA National Geophysical Data Center. ETOPO1 1 Arc-Minute Global Relief Model NOAA National Centers for Environmental Information2009. Available from: https://www.ncei.noaa.gov/access/metadata/landing-page/bin/iso?id=gov.noaa.ngdc.mgg.dem:316.

56. Knopp S, Mohammed KA, Ali SM, Khamis IS, Ame SM, Albonico M, et al. Study and implementation of urogenital schistosomiasis elimination in Zanzibar (Unguja and Pemba islands) using an integrated multidisciplinary approach. BMC Public Health. 2012;12:930. doi: https://doi.org/10.1186/1471-2458-12-930.

57. Knopp S, Ame SM, Person B, Hattendorf J, Rabone M, Juma S, et al. A 5-year intervention study on elimination of urogenital schistosomiasis in Zanzibar: Parasitological results of annual cross-sectional surveys. PLoS Negl Trop Dis. 2019;13(5). doi: https://doi.org/10.1371/journal.pntd.0007268.

58. Knopp S, Person B, Ame SM, Ali SM, Hattendorf J, Juma S, et al. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: A cluster-randomised trial. The Lancet Global Health. 2019;7(8):e1118-e29. doi: https://doi.org/10.1016/S2214-109X(19)30189-5.

59. Trippler L, Ame SM, Hattendorf J, Juma S, Abubakar S, Ali SM, et al. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: Repeated cross-sectional studies. PLoS Negl Trop Dis. 2021;15(2):e0009127. doi: https://doi.org/10.1371/journal.pntd.0009127.

60. Trippler L, Hattendorf J, Ali SM, Ame SM, Juma S, Kabole F, et al. Novel tools and strategies for breaking schistosomiasis transmission: Study protocol for an intervention study. BMC Infect Dis. 2021;21(1):1024. doi: https://doi.org/10.1186/s12879-021-06620-8.

61. World Health Organization. Schistosomiasis progress report 2001-2011 and strategic plan 2012-2020. World Health Organization, 2013. Available from: https://apps.who.int/iris/handle/10665/78074

62. Abudho BO, Ndombi EM, Guya B, Carter JM, Riner DK, Kittur N, et al. Impact of four years of annual mass drug administration on prevalence and intensity of schistosomiasis among primary and high school children in western Kenya: A repeated cross-sectional study. Am J Trop Med Hyg. 2018;98(5):1397-402. doi: https://doi.org/10.4269/ajtmh.17-0908.

63. Kura K, Ayabina D, Hollingsworth TD, Anderson RM. Determining the optimal strategies to achieve elimination of transmission for *Schistosoma mansoni*. Parasit Vectors. 2022;15(1):55. doi: https://doi.org/10.1186/s13071-022-05178-x.

64. Kura K, Hardwick RJ, Truscott JE, Toor J, Hollingsworth TD, Anderson RM. The impact of mass drug administration on *Schistosoma haematobium* infection: what is required to achieve morbidity control and elimination? Parasit Vectors. 2020;13(1):554. doi: https://doi.org/10.1186/s13071-020-04409-3.

65. Secor WE, Wiegand RE, Montgomery SP, Karanja DMS, Odiere MR. Comparison of school-based and community-wide mass drug administration for schistosomiasis control in an area of western Kenya with high initial *Schistosoma mansoni* infection prevalence: A cluster randomized trial. Am J Trop Med Hyg. 2020;102(2):318-27. doi: https://doi.org/10.4269/ajtmh.19-0626.

66. Gurarie D, Lo NC, Ndeffo-Mbah ML, Durham DP, King CH. The human-snail transmission environment shapes long term schistosomiasis control outcomes: Implications for improving the accuracy of predictive modeling. PLoS Negl Trop Dis. 2018;12(5):e0006514. doi: https://doi.org/10.1371/journal.pntd.0006514.

67. Morozoff C, Avokpaho E, Puthupalayam Kaliappan S, Simwanza J, Gideon SP, Lungu W, et al. Costs of community-wide mass drug administration and school-based deworming for soil-transmitted helminths: evidence from a randomised controlled trial in Benin, India and Malawi. BMJ Open. 2022;12(7):e059565. doi: https://doi.org/10.1136/bmjopen-2021-059565.

68. Lo NC, Bogoch, II, Blackburn BG, Raso G, N'Goran EK, Coulibaly JT, et al. Comparison of community-wide, integrated mass drug administration strategies for schistosomiasis and soil-transmitted helminthiasis: a cost-effectiveness modelling study. Lancet Glob Health. 2015;3(10):e629-38. doi: https://doi.org/10.1016/s2214-109x(15)00047-9.

69. Mduluza-Jokonya TL, Naicker T, Jokonya L, Midzi H, Vengesai A, Kasambala M, et al. Association of *Schistosoma haematobium* infection morbidity and severity on co-infections

in pre-school age children living in a rural endemic area in Zimbabwe. BMC Public Health. 2020;20(1):1570. doi: https://doi.org/10.1186/s12889-020-09634-0.

70. Masaku J, Njomo DW, Njoka A, Okoyo C, Mutungi FM, Njenga SM. Soil-transmitted helminths and schistosomiasis among pre-school age children in a rural setting of Busia County, Western Kenya: a cross-sectional study of prevalence, and associated exposures. BMC Public Health. 2020;20(1):356. doi: https://doi.org/10.1186/s12889-020-08485-z.

71. Isaiah PM, Palmeirim MS, Steinmann P. Epidemiology of pediatric schistosomiasis in hard-to-reach areas and populations: A scoping review protocol2022. doi: https://doi.org/10.12688/f1000research.126884.2.

72. Pediatric Praziquantel Consortium. 2023 [26.07.2023]. Available from: https://www.pediatricpraziquantelconsortium.org/.

73. Mansfield-Aders W. Annual report of the biological division for 1927. Wellcome Collection: 1928.

74. Macdonald G, Forsyth DM, Rayski C, Rashid C. The control of urinary schistosomiasis. Trials of niridazole and other drugs in children attending primary schools in Zanzibar, Tanzania, and the failure of a pilot eradication programme. Annals of tropical medicine and parasitology. 1968;62(4):407-15. doi: https://doi.org/10.1080/00034983.1968.11686577.

75. Sokolow SH, Wood CL, Jones IJ, Swartz SJ, Lopez M, Hsieh MH, et al. Global assessment of schistosomiasis control over the past century shows targeting the snail intermediate host works best. PLoS Negl Trop Dis. 2016;10(7):e0004794. doi: https://doi.org/10.1371/journal.pntd.0004794.

76. Sturrock R, Barnish G, Upatham ES. Snail findings from an experimental molluscicide programme to control *Schistosoma mansoni* transmission on St. Lucia. Int J Parasitol. 1974;4(3):231-40. doi: https://doi.org/10.1016/0020-7519(74)90079-4.

77. Lo NC, Gurarie D, Yoon N, Coulibaly JT, Bendavid E, Andrews JR, et al. Impact and cost-effectiveness of snail control to achieve disease control targets for schistosomiasis. Proc Natl Acad Sci U S A. 2018;115(4):E584-E91. doi: https://doi.org/10.1073/pnas.1708729114.

78. Mozley A. XXVI.—The fresh-water mollusca of the Tanganyika territory and Zanzibar Protectorate, and their relation to human schistosomiasis. Transactions of the Royal Society of Edinburgh. 1939;59(3):687-744. doi: https://doi.org/10.1017/S0080456800017403.

79. El-Seedi HR, Khalifa SAM, Mohamed AH, Yosri N, Zhao C, El-Wakeil N, et al. Plant extracts and compounds for combating schistosomiasis. Phytochemistry Reviews. 2022. doi: https://doi.org/10.1007/s11101-022-09836-x.

80. Rohr JR, Sack A, Bakhoum S, Barrett CB, Lopez-Carr D, Chamberlin AJ, et al. A planetary health innovation for disease, food and water challenges in Africa. Nature. 2023;619(7971):782-7. doi: https://doi.org/10.1038/s41586-023-06313-z.

81. Person B, Ali SM, A'Kadir FM, Ali JN, Mohammed UA, Mohammed KA, et al. Community knowledge, perceptions, and practices associated with urogenital schistosomiasis among school-aged children in Zanzibar, United Republic of Tanzania. PLoS Negl Trop Dis. 2016;10(7). doi: https://doi.org/10.1371/journal.pntd.0004814.

82. Person B, Rollinson D, Ali SM, Mohammed UA, A'Kadir F M, Kabole F, et al. Evaluation of a urogenital schistosomiasis behavioural intervention among students from rural schools in Unguja and Pemba islands, Zanzibar. Acta Trop. 2021;220:105960. doi: https://doi.org/10.1016/j.actatropica.2021.105960.

83. Lar LA, Dean L, Adekeye T, Oluwole AS, Lawong D, Kevin G, et al. Lessons from participatory community mapping to inform neglected tropical disease programmes in Nigeria. Int Health. 2023;15(Suppl 1):i6-i17. doi: https://doi.org/10.1093/inthealth/ihac074.

84. Adam I, A. AN, Al-Wutayd O, Khamis AH. Prevalence of schistosomiasis and its association with anemia among pregnant women: a systematic review and meta-analysis. Parasit Vectors. 2021;14(1):133. doi: https://doi.org/10.1186/s13071-021-04642-4.

85. Salawu OT, Odaibo AB. Maternal schistosomiasis: A growing concern in sub-Saharan Africa. Pathog Glob Health. 2014;108(6):263-70. doi: https://doi.org/10.1179/2047773214y.0000000150.

86. Young MF. Maternal anaemia and risk of mortality: a call for action. Lancet Glob Health. 2018;6(5):e479-e80. doi: https://doi.org/10.1016/s2214-109x(18)30185-2.

87. Rahmati S, Delpishe A, Azami M, Hafezi Ahmadi MR, Sayehmiri K. Maternal anemia during pregnancy and infant low birth weight: A systematic review and meta-analysis. Int J Reprod Biomed. 2017;15(3):125-34. doi: https://doi.org/10.29252/ijrm.15.3.125.

88. Ondigo BN, Muok EMO, Oguso JK, Njenga SM, Kanyi HM, Ndombi EM, et al. Impact of mothers' schistosomiasis status during gestation on children's IgG antibody responses to

routine vaccines 2 years later and anti-schistosome and anti-malarial responses by neonates in western Kenya. Front Immunol. 2018;9:1402. doi: https://doi.org/10.3389/fimmu.2018.01402.

89. Person B, Knopp S, Ali SM, A'Kadir FM, Khamis AN, Ali JN, et al. Community co-designed schistosomiasis control interventions for school-aged children in Zanzibar. Journal of biosocial science. 2016;48:S56-S73. doi: https://doi.org/10.1017/S0021932016000067.

90. Sousa-Figueiredo JC, Stanton MC, Katokele S, Arinaitwe M, Adriko M, Balfour L, et al. Mapping of schistosomiasis and soil-transmitted helminths in Namibia: The first large-scale protocol to formally include rapid diagnostic tests. PLoS Negl Trop Dis. 2015;9(7):e0003831. doi: https://doi.org/10.1371/journal.pntd.0003831.

91. Meginnis K, Hanley N, Mujumbusi L, Lamberton PHL. Non-monetary numeraires: Varying the payment vehicle in a choice experiment for health interventions in Uganda. Ecol Econ. 2020;170:106569. doi: https://doi.org/10.1016/j.ecolecon.2019.106569.

92. Schur N, Hürlimann E, Stensgaard A-S, Chimfwembe K, Mushinge G, Simoonga C, et al. Spatially explicit *Schistosoma* infection risk in eastern Africa using Bayesian geostatistical modelling. Acta Trop. 2013;128(2):365-77. doi: https://doi.org/10.1016/j.actatropica.2011.10.006.

93. Trippler L, Ali SM, Ame SM, Hattendorf J, Suleiman KR, Ali MN, et al. Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: Baseline cross-sectional survey findings before the onset of a 3-year intervention study. Parasit Vectors. 2022;15(1):292. doi: https://doi.org/10.1186/s13071-022-05404-6.

94. Trippler L, Ali MN, Ame SM, Ali SM, Kabole F, Hattendorf J, et al. GPS-based fine-scale mapping surveys for schistosomiasis assessment: A practical introduction and documentation of field implementation. Infect Dis Poverty. 2022;11(1):8. doi: https://doi.org/10.1186/s40249-021-00928-y.

95. Kepha S, Ochol D, Wakesho F, Omondi W, Njenga SM, Njaanake K, et al. Precision mapping of schistosomiasis and soil-transmitted helminthiasis among school age children at the coastal region, Kenya. PLoS Negl Trop Dis. 2023;17(1):e0011043. doi: https://doi.org/10.1371/journal.pntd.0011043.

96. Phillips AE, Ower AK, Mekete K, Liyew EF, Maddren R, Belay H, et al. Association between water, sanitation, and hygiene access and the prevalence of soil-transmitted helminth

and schistosome infections in Wolayita, Ethiopia. Parasit Vectors. 2022;15(1):410. doi: https://doi.org/10.1186/s13071-022-05465-7.

97. Wiegand RE, Fleming FM, de Vlas SJ, Odiere MR, Kinung'hi S, King CH, et al. Defining elimination as a public health problem for schistosomiasis control programmes: Beyond prevalence of heavy-intensity infections. The Lancet Global Health. 2022;10(9):e1355-e9. doi: https://doi.org/10.1016/S2214-109X(22)00287-X.

98. Pennance T, Person B, Muhsin MA, Khamis AN, Muhsin J, Khamis IS, et al. Urogenital schistosomiasis transmission on Unguja Island, Zanzibar: Characterisation of persistent hot-spots. Parasit Vectors. 2016;9(1):646. doi: https://doi.org/10.1186/s13071-016-1847-0.

99. Nwoko OE, Manyangadze T, Chimbari MJ. Spatial and seasonal distribution of human schistosomiasis intermediate host snails and their interactions with other freshwater snails in 7 districts of KwaZulu-Natal province, South Africa. Sci Rep. 2023;13(1):7845. doi: https://doi.org/10.1038/s41598-023-34122-x.

100. Kane RA, Stothard JR, Emery AM, Rollinson D. Molecular characterization of freshwater snails in the genus *Bulinus*: a role for barcodes? Parasit Vectors. 2008;1(1):15. doi: https://doi.org/10.1186/1756-3305-1-15.

101. Kelly-Hope LA, Unnasch TR, Stanton MC, Molyneux DH. Hypo-endemic onchocerciasis hotspots: Defining areas of high risk through micro-mapping and environmental delineation. Infect Dis Poverty. 2015;4:36. doi: https://doi.org/10.1186/s40249-015-0069-6.

102. Hardy A, Ettritch G, Cross DE, Bunting P, Liywalii F, Sakala J, et al. Automatic detection of open and vegetated water bodies using sentinel 1 to map African malaria vector mosquito breeding habitats. Remote Sensing. 2019;11(5):593. doi: https://doi.org/10.3390/rs11050593.

103. Simoonga C, Utzinger J, Brooker S, Vounatsou P, Appleton CC, Stensgaard AS, et al. Remote sensing, geographical information system and spatial analysis for schistosomiasis epidemiology and ecology in Africa. Parasitology. 2009;136(13):1683-93. doi: https://doi.org/10.1017/S0031182009006222.

104. Read JM, Chambers C, Torrado M. Remote sensing. In: Kobayashi A, editor. International Encyclopedia of Human Geography (Second Edition). Oxford: Elsevier; 2020. p. 411-22. doi: https://doi.org/10.1016/B978-0-08-102295-5.10589-X.

105. Braae UC, Magnussen P, Ndawi B, Harrison W, Lekule F, Johansen MV. Effect of repeated mass drug administration with praziquantel and track and treat of taeniosis cases on the prevalence of taeniosis in *Taenia solium* endemic rural communities of Tanzania. Acta Trop. 2017;165:246-51. doi: https://doi.org/10.1016/j.actatropica.2015.10.012.

106. Celone M, Person B, Ali SM, Lyimo JH, Mohammed UA, Khamis AN, et al. Increasing the reach: Involving local Muslim religious teachers in a behavioral intervention to eliminate urogenital schistosomiasis in Zanzibar. Acta Trop. 2016;163:142-8. doi: https://doi.org/10.1016/j.actatropica.2016.08.004.

107. Vilakati S, Mngadi N, Benjamin-Chung J, Dlamini N, Dufour M-SK, Whittemore B, et al. Effectiveness and safety of reactive focal mass drug administration (rfMDA) using dihydroartemisinin–piperaquine to reduce malaria transmission in the very low-endemic setting of Eswatini: A pragmatic cluster randomised controlled trial. BMJ Global Health. 2021;6(6):e005021. doi: https://doi.org/10.1136/bmjgh-2021-005021.

108. King CH, Bertsch D. Historical perspective: snail control to prevent schistosomiasis. PLoS Negl Trop Dis. 2015;9(4):e0003657. doi: https://doi.org/10.1371/journal.pntd.0003657.

109. Huang Q, Gurarie D, Ndeffo-Mbah M, Li E, King CH. *Schistosoma* transmission in a dynamic seasonal environment and its impact on the effectiveness of disease control. J Infect Dis. 2020;225(6):1050-61. doi: https://doi.org/10.1093/infdis/jiaa746.

110. Augusto G, Magnussen P, Kristensen TK, Appleton CC, Vennervald BJ. The influence of transmission season on parasitological cure rates and intensity of infection after praziquantel treatment of *Schistosoma haematobium*-infected schoolchildren in Mozambique. Parasitology. 2009;136(13):1771-9. doi: https://doi.org/10.1017/S0031182009006210.

APPENDICES

Appendix A: Publication "The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones"

Lydia Trippler[1,2], Stefanie Knopp[1,2], Sophie Welsche[3], Bonnie L. Webster[4,5], J. Russell Stothard[6], Lynsey Blair[3], Fiona Allan[4,5,7], Shaali Makame Ame[8], Saleh Juma[9], Fatma Kabole[8], Said Mohammed Ali[10], David Rollinson[4,5,11], Tom Pennance[4,5,12]

1 Swiss Tropical and Public Health Institute, Allschwil, Switzerland

2 University of Basel, Basel, Switzerland

3 Unlimit Health, London, United Kingdom

4 Natural History Museum, London, United Kingdom

5 London Centre for Neglected Tropical Disease Research, London, United Kingdom

6 Liverpool School of Tropical Medicine, Liverpool, United Kingdom

7 University of St Andrews, St Andrews, United Kingdom

8 Neglected Diseases Programme, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania

9 Neglected Diseases Programme, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania

10 Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania

11 Global Schistosomiasis Alliance, London, United Kingdom

12 Western University of Health Sciences, Lebanon, Oregon, United States of America

CHAPTER TWO

The long road to schistosomiasis elimination in Zanzibar: A systematic review covering 100 years of research, interventions and control milestones

Lydia Trippler[a,b,*], Stefanie Knopp[a,b], Sophie Welsche[c], Bonnie L. Webster[d,e], J. Russell Stothard[f], Lynsey Blair[c], Fiona Allan[d,e,g], Shaali Makame Ame[h], Saleh Juma[i], Fatma Kabole[h], Said Mohammed Ali[j], David Rollinson[d,e,k], and Tom Pennance[d,e,l,*]

[a]Swiss Tropical and Public Health Institute, Allschwil, Switzerland
[b]University of Basel, Basel, Switzerland
[c]Unlimit Health, London, United Kingdom
[d]Natural History Museum, London, United Kingdom
[e]London Centre for Neglected Tropical Disease Research, London, United Kingdom
[f]Liverpool School of Tropical Medicine, Liverpool, United Kingdom
[g]University of St Andrews, St Andrews, United Kingdom
[h]Neglected Diseases Programme, Zanzibar Ministry of Health, Lumumba, Unguja, United Republic of Tanzania
[i]Neglected Diseases Programme, Zanzibar Ministry of Health, Mkoroshoni, Pemba, United Republic of Tanzania
[j]Public Health Laboratory – Ivo de Carneri, Wawi, Chake Chake, Pemba, United Republic of Tanzania
[k]Global Schistosomiasis Alliance, London, United Kingdom
[l]Western University of Health Sciences, Lebanon, OR, United States
*Corresponding authors. e-mail address: lydia.trippler@swisstph.ch; t.pennance@nhm.ac.uk

Contents

1. Introduction 74
2. Methods 77
   2.1 Search strategy 77
   2.2 Inclusion and exclusion criteria 78
   2.3 Process of including and excluding records 78
   2.4 Writing the sections 79
3. Results of records search 79
4. Overview of the schistosomiasis control, research and involvement of international stakeholders across the past century 80
5. Epidemiology of schistosomiasis in Zanzibar 91
   5.1 *Schistosoma* spp. on Unguja and Pemba 91
   5.2 *Schistosoma haematobium* prevalence changes over 100 years 92
   5.3 Spatial heterogeneity of *Schistosoma haematobium* infections in Zanzibar 95

Advances in Parasitology, Volume 122
ISSN 0065-308X, https://doi.org/10.1016/bs.apar.2023.06.001

5.4 Sex-related difference in *Schistosoma haematobium* prevalence 96
5.5 Age-related difference in *Schistosoma haematobium* prevalence 98
5.6 Outlook 99
6. Biology of the intermediate host snails 99
6.1 Endemic *Bulinus* species of Unguja and Pemba 99
6.2 Taxonomic history and identification of *Bulinus globosus* and *Bulinus nasutus* 100
6.3 Incrimination of *Bulinus* as intermediate host(s) of *Schistosoma haematobium* 105
6.4 Incrimination of *Bulinus* as intermediate host(s) of *Schistosoma bovis* 109
6.5 Monitoring of *Schistosoma* infections in *Bulinus* spp 109
6.6 Habitat preferences of *Bulinus globosus* and *Bulinus nasutus* 110
6.7 Interspecies and intraspecies genetic diversity of *Bulinus globosus* and *Bulinus nasutus* 112
6.8 Outlook 113
7. Molecular epidemiology 115
7.1 Genetic epidemiology of *Schistosoma haematobium* in Zanzibar 115
7.2 Genetic diversity and population structure of *Schistosoma haematobium* on Unguja and Pemba 116
7.3 Do intense control interventions change the composition of *Schistosoma haematobium* populations? 118
7.4 Emergence of *Schistosoma bovis* in Zanzibar 119
7.5 Outlook 120
8. Diagnostics 121
8.1 First diagnostic research in Zanzibar 121
8.2 Immuno-diagnosis-antibody detection 122
8.3 Haematuria-based diagnosis 125
8.4 Using haematuria as a proxy for treatment and sign of morbidity reduction 126
8.5 Urine analysis and urology as a diagnostic and morbidity marker 130
8.6 Immuno-diagnosis; antigen detection 132
8.7 Molecular (DNA) diagnostics 133
8.8 Outlook 135
9. Morbidity 136
9.1 Radiological abnormalities discovered with urograms 136
9.2 Exceptional case report findings 139
9.3 Uropathy discovered with ultrasonography 140
9.4 Anthropometric measures and anaemia 142
9.5 Outlook 143
10. Treatment interventions 144
10.1 Early antischistosomal treatments in Zanzibar 144
10.2 Early test-and-treat approaches and community-wide selective chemotherapy 146
10.3 National Helminth Control Programme: Preventive chemotherapy for morbidity control 149

10.4 Establishment of a national schistosomiasis control programme and integrated MDA 151
10.5 Aiming for elimination of urogenital schistosomiasis: The national plan and moving beyond MDA 152
10.6 Outlook 154
11. Snail control interventions 155
11.1 Snail control interventions in the 20th century 155
11.2 Large-scale snail control interventions 158
11.3 Outlook 161
12. Behaviour change interventions 162
12.1 From health education to behaviour change interventions 162
12.2 Large-scale behaviour change interventions 164
12.3 Outlook 167
13. Surveillance-response interventions 168
13.1 Implementation of surveillance-response interventions in Zanzibar 168
13.2 Outlook 170
14. Conclusion 171
Acknowledgements 175
Disclosure 175
Appendix A Supporting information 175
References 176

Abstract

Zanzibar is among the few places in sub-Saharan Africa where interruption of *Schistosoma* transmission seems an achievable goal. Our systematic review identifies and discusses milestones in schistosomiasis research, control and elimination efforts in Zanzibar over the past 100 years.

The search in online databases, libraries, and the World Health Organization Archives revealed 153 records published between May 1928 and August 2022. The content of records was summarised to highlight the pivotal work leading towards urogenital schistosomiasis elimination and remaining research gaps.

The greatest achievement following 100 years of schistosomiasis interventions and research is undoubtedly the improved health of Zanzibaris, exemplified by the reduction in *Schistosoma haematobium* prevalence from > 50% historically down to < 5% in 2020, and the absence of severe morbidities. Experiences from Zanzibar have contributed to global schistosomiasis guidelines, whilst also revealing challenges that impede progression towards elimination. Challenges include: transmission heterogeneity requiring micro-targeting of interventions, post-treatment recrudescence of infections in transmission hotspots, biological complexity of intermediate host snails, emergence of livestock *Schistosoma* species complicating surveillance whilst creating the risk for interspecies hybridisation, insufficient diagnostics performance for light intensity infections and female genital schistosomiasis, and a lack of acceptable sanitary alternatives to freshwater bodies.

Our analysis of the past revealed that much can be achieved in the future with practical implementation of integrated interventions, alongside operational research. With continuing national and international commitments, interruption of *S. haematobium* transmission across both islands is within reach by 2030, signposting the future demise of urogenital schistosomiasis across other parts of sub-Saharan Africa.

Abbreviations

CAA	circulating anodic antigen.
CCA	circulating cathodic antigen.
DBL	Danish Bilharziasis Laboratory.
eDNA	environmental DNA.
ELISA	enzyme-linked immunosorbent assay.
FGS	female genital schistosomiasis.
HWCS	human water contact site.
IHA	indirect haemagglutination.
ITS	internal transcribed spacer.
***cox*1**	cytochrome oxidase subunit 1.
***nad*1**	NADH dehydrogenase subunit 1.
MDA	mass drug administration.
MGS	male genital schistosomiasis.
MoH	Ministry of Health.
NHM	Natural History Museum of London.
NTD	neglected tropical disease.
PHL-IdC	Public Health Laboratory-Ivo de Carneri.
RAPD	random amplified polymorphic DNA.
RDT	rapid diagnostic test.
RFLP	restriction fragment length polymorphism.
RPA	recombinase polymerase amplification.
SCI	Schistosomiasis Control Initiative.
SCORE	Schistosomiasis Consortium for Operational Research and Evaluation.
SEA	soluble egg antigen.
Swiss TPH	Swiss Tropical and Public Health Institute.
UACR	urine albumin-to-creatinine ratio.
UCP-LF	up-converting phosphor-lateral flow.
WHO	World Health Organization.
ZEST	Zanzibar elimination of schistosomiasis.

1. Introduction

Human schistosomiasis is a neglected tropical disease (NTD) that poses a risk to over 700 million people worldwide (World Health Organization, 2023a). At least 251.4 million people requiring preventive chemotherapy when last estimated in 2021, of which only ~30% likely received treatment. Nearly 90% of *Schistosoma* spp. infected individuals in

need of treatment for intestinal and urogenital schistosomiasis reside in sub-Saharan Africa (World Health Organization, 2020). Urogenital schistosomiasis is caused by the parasitic flatworm *S. haematobium*, one of the nine currently described species within the *Schistosoma haematobium* species group that all utilise snails of the genus *Bulinus* as intermediate hosts in their life cycle (Brown, 1994, Webster et al., 2006, Hanelt et al., 2009, Colley et al., 2014). *Schistosoma haematobium* is widely distributed across Africa, Madagascar, the Middle East and cases have also been reported from Corsica (France) (Boissier et al., 2016). Pathogenesis and morbidity in the definitive human host is induced by eggs which become trapped in surrounding tissues, specifically the bladder, kidneys and reproductive organs leading to granuloma formation and chronic inflammation that can lead to organ failure, squamous cell carcinomas and death (Colley et al., 2014). The morbidity associated with the reproductive organs [female genital schistosomiasis (FGS) and male genital schistosomiasis (MGS)] also manifests as stigmatising symptoms, including a heightened risk of transmission of the human immunodeficiency virus and fertility issues (Kayuni et al., 2019, Sturt et al., 2020, Bustinduy et al., 2022).

One region that used to be highly endemic for urogenital schistosomiasis in sub-Saharan Africa in the last century is the Zanzibar Archipelago, United Republic of Tanzania. Over the past decades, however, infections and morbidity were substantially reduced on the two main islands, Unguja and Pemba. Today, urogenital schistosomiasis has been successfully eliminated as a public health problem from most areas of Zanzibar. Current goals are therefore targeted towards reducing the incidence of new infections to zero, which could make Zanzibar one of the first regions in sub-Saharan Africa to achieve interruption of *Schistosoma* transmission.

Situated off the east coast of Tanzania, separated from the African mainland by a channel of the Indian Ocean, Unguja and Pemba have been important ports for trade routes between the Arabian Peninsula and the African continent, possibly as early as the late 5th to 8th century Common Era (Crowther et al., 2015). During the past millennium, Zanzibar fell under the control of Portuguese, Omani and then British colonisers, but shortly after the revolution in early 1964 the islands were liberated from British colonial rule, which led to the unification of Zanzibar with Tanganyika (Tanzania mainland) forming the United Republic of Tanzania (Office of Chief Government Statistician Zanzibar, 2020). However, the islands, with their own unique Swahili-Arabic culture, acquired from a blend of the islands' rich history and the natural environment, remain a

semi-autonomous region with a population of ~1.9 million inhabitants in 2022 (Ministry of Finance and Planning - National Bureau of Statistics Tanzania and President's Office - Finance and Planning - Office of the Chief Government Statistician Zanzibar, 2022).

The geographical separation of Unguja and Pemba from mainland Africa, coupled with the islands' degree of self-government, has made the islands a microcosm with its own public health policies. This microcosm has led to the identification of Unguja and Pemba as 'model islands' for the implementation and evaluation of the effectiveness of several programmes for the control and elimination of infectious diseases in sub-Saharan Africa, including malaria, trypanosomiasis and lymphatic filariasis (Vreysen et al., 2000, Mohammed et al., 2006, Bhattarai et al., 2007, Aregawi et al., 2011, Rebollo et al., 2015). Infectious disease control programmes have benefited from Unguja and Pemba being divided into 5 regions, 11 districts and 388 small administrative areas, called shehias (Office of Chief Government Statistician Zanzibar, 2020, Ministry of Health (MoH) [Tanzania Mainland], 2023), which allows control programmes to be implementable at different population and geographic levels. Just as with malaria, trypanosomiasis and lymphatic filariasis, collective control efforts spanning almost a century (which are summarised in this review) have alleviated much of the severe morbidity and mortality associated with *S. haematobium* infections on Unguja and Pemba through preventive chemotherapy, and other interventions targeted at reducing parasite transmission. In terms of achieving the goals set by the World Health Organization (WHO) for the interruption of *Schistosoma* transmission in humans in selected countries by 2030, the islands of Zanzibar are perhaps the closest in sub-Saharan Africa at being able to do so (World Health Organization, 2013, 2023b). However, interruption of *Schistosoma* transmission has not yet been achieved and transmission persists in some regions. This highlights the challenges for the interruption of *Schistosoma* transmission in environments where snail intermediate hosts thrive and no adequate infrastructure exists to offer an alternative to direct human water contact with freshwater transmission sites. Additionally, the risk of resurgence of transmission remains real if sustained efforts cannot be maintained, even after years of control and interventions that have kept infections levels low.

The following systematic review aims to identify and discuss the milestones in schistosomiasis research, control and elimination efforts over the past 100 years on Unguja and Pemba. The work conducted, and achievements made, are discussed across key themes encompassing

Schistosoma transmission and control, namely: disease epidemiology, biology and control of the intermediate host snails, molecular epidemiology of *Schistosoma* populations, diagnostics, morbidity assessments and treatment, behaviour change and surveillance-response interventions. This review highlights the importance that work on these islands has had in shaping current WHO schistosomiasis control and elimination policies and guidelines. Now, the focus of future interventions on Unguja and Pemba will be on the complete interruption of *Schistosoma* transmission (zero incidence) and on how to quickly detect and counteract resurgence through appropriate surveillance (WHO, 2022). With this in mind, the following systematic review highlights the challenges of schistosomiasis control and elimination and concludes with recommendations for future research and interventions that may be required for achieving interruption of *Schistosoma* transmission in humans by 2030, in Zanzibar and also other settings nearing elimination.

2. Methods

2.1 Search strategy

This systematic review has been conducted following the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) statement (Page et al., 2021). A comprehensive search for literature within four different online databases (PubMed, Embase, SCOPUS, and Web of Science) was initiated to retrieve records published until and including the 17th August 2022. The online search strategy was designed based on two key concepts: (1) 'Unguja', 'Pemba', 'Zanzibar', and (2) 'Bilharz*', '*Bulinus*', 'Praziquantel', '*Schistosom**', 'Snail'. All keywords were connected by Boolean operators.

In addition to the online database search, the WHO Archives were contacted by email to identify additional records on schistosomiasis and associated snail research in Zanzibar. Once access to the WHO Archives was granted, records were retrieved electronically (13th September 2022) or viewed on site at the WHO Archives in Geneva, Switzerland (3rd November 2022).

Furthermore, a literature search about schistosomiasis and related snail research was conducted on the Swiss Library Service Platform (www.swisscovery.slsp.ch) with a subsequent visit at the library of the Swiss Tropical and Public Health Institute (Swiss TPH) in Allschwil, Switzerland

(17th August 2023). Known British protectorate reports for Zanzibar were received from the Wellcome Collection (9th November 2022) and the London School of Economics (10th December 2019), United Kingdom. Finally, additional records identified in the bibliography of identified records, were retrieved through a targeted online search.

2.2 Inclusion and exclusion criteria

All records published in the respective sources up to the date of the search were retrieved for full-text screening if the topic of schistosomiasis or related snail research on the Zanzibar islands was covered by the title or abstract. Records that were not related to schistosomiasis research conducted on the Zanzibar islands, or schistosomiasis research conducted outside the Zanzibar islands in the full text, were excluded from the review. In addition, records were excluded from the review if they fell into one of the following groups: (1) not related to the topic of the review after full-text screening, (2) conference abstracts where the same results were later published in a scientific paper, or (3) originals of Corrigenda.

2.3 Process of including and excluding records

Titles and abstracts of all records retrieved from the online database search and all records retrieved electronically from the WHO Archives were screened for schistosomiasis and related snail research in Zanzibar by two independent authors (LT and TP). As only one author (LT) was able to visit the WHO Archives and the library of the Swiss TPH in person, LT performed the initial reference screening for onsite retrieval alone. All records that were only available on site and that LT considered important were scanned by LT and made available electronically to TP. As only one person (TP) was able to visit the Wellcome Collection and the London School of Economics in person, TP performed the initial record screening for onsite retrieval alone. All records that were only available on site and that TP considered important were scanned by TP and made available electronically to LT. For all the collected records, LT and TP subsequently screened the records independently and decided on their inclusion or exclusion. In case of disagreement about including the records into the review, LT and TP reread the records and discussed whether the inclusion criteria were met until a consensus was reached.

Finally, two authors (LT and TP) classified all records that were selected for inclusion in the review into at least one of the following topics: (1) International stakeholders in schistosomiasis control and research, (2) Epidemiology of schistosomiasis in Zanzibar, (3) Biology of the intermediate

host snail, (4) Molecular epidemiology, (5) Diagnostics, (6) Morbidity, (7) Treatment interventions, (8) Snail control interventions, (9) Behaviour change interventions, (10) Surveillance-response interventions.

2.4 Writing the sections

Each topic led to a separate section of the review and was written by at least two authors according to the following scheme: (1) reading all studies and reports assigned to the main topic of the section, (2) summarising the approaches, interventions, research, and results described in the respective studies and reports, (3) discussing the implications derived for Zanzibar and elsewhere, and (4) discussing open research questions that need to be addressed in the future to achieve interruption of *S. haematobium* transmission. In addition, each author provided key findings and research needs, based on which the introduction and conclusion of the review was written.

3. Results of records search

As a result of the online database search, 392 records were retrieved, of which 261 (66.6%) were removed as duplicates and 131 (33.4%) were included in the initial database screening (Fig. 1). During the records

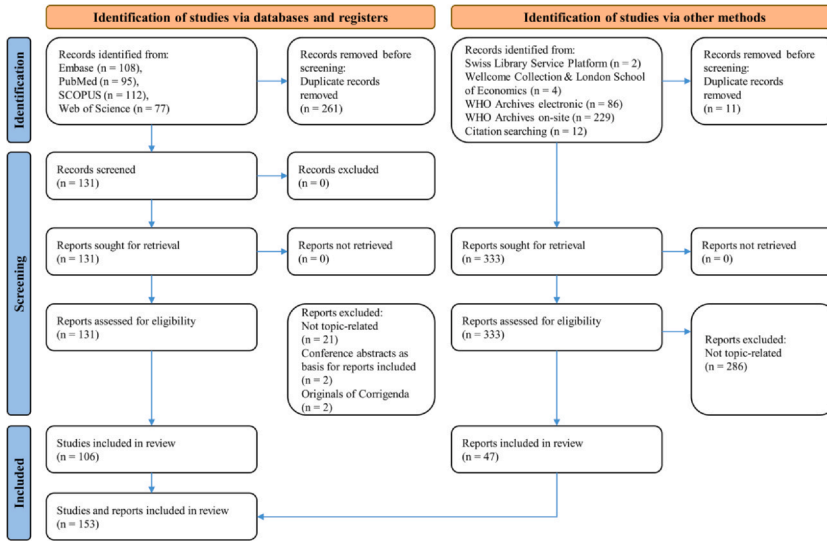


Fig. 1 Flow diagram of the results of the systematic search of the literature for schistosomiasis and related snail research in Zanzibar. *Adapted from Page et al. (2021).*

screening, 25 (18.3%) records were excluded as they were either: (1) not related to the topic of the review after full-text screening, (2) conference abstracts where the same results were later published in a scientific paper, or (3) originals of Corrigenda. Finally, 106 studies were included in the review.

Two records were identified through the Swiss Library Service Platform and four through the Wellcome Collection, and London School of Economics (Fig. 1). An additional 86 and 229 records were retrieved from the WHO Archives electronically and onsite, respectively. Finally, 12 records were identified through citations provided in records originally retrieved from the online database search or from the WHO Archives. In total, 333 records were identified via these 'other methods', of which 11 records were removed as duplicates. During the records screening, 286 records were excluded because they were not relevant to the topic of the review.

In total, 106 studies [as defined in the PRISMA 2020 statement (Page et al., 2021)] from the online database search and 47 reports from 'other methods' were included in the review. These 153 studies and reports covered a period from 1927 to 2022 (Supplementary File 1: Table 1, and Fig. 2).

4. Overview of the schistosomiasis control, research and involvement of international stakeholders across the past century

The review will provide a very detailed account of the schistosomiasis activities undertaken during the last century. To better understand the timeline of research, project implementation and international partnerships with the Zanzibar Ministry of Health (MoH) over the past 100 years, an overview is provided here of the governmental bodies, research institutes, charitable organisations and companies involved.

The first record of urogenital schistosomiasis infection on Unguja and Pemba dates back to 1925, when 138 individuals were treated in the British Colonial Government hospital (Semple, 1927). It should be noted that although the British reports about the Zanzibar Protectorate that are detailed in this review (Semple, 1927, Mansfield-Aders, 1928, Zanzibar Protectorate, 1930–1934) contain useful information regarding urogenital schistosomiasis on Unguja and Pemba, they reflect colonialistic views and contain descriptions that go against today's ethical standards. During the early time points of the 1920s, few individuals were treated, however, it is

Table 1 Milestones and research programmes contributing to schistosomiasis research and control on the Zanzibar islands. CCA = Circulating Cathodic Antigen, MDA = Mass Drug Administration, WHO = World Health Organization, ZEST = Zanzibar Elimination of Schistosomiasis Transmission.

Year	Milestone	Outcome	Reference (s)
1925	First records of urogenital schistosomiasis treatment in Zanzibar.	Urogenital schistosomiasis recognised as a public health problem in Zanzibar.	Zanzibar Protectorate (1930–1934)
1926	Cawston & Mansfield-Aders lead first research into schistosomiasis in Zanzibar.	Identify snail intermediate host and regions of transmission.	Cawston (1927b), Zanzibar Protectorate (1930-1934)
1927	First report of a diagnostic test to detect *S. haematobium* infection in humans (centrifugation of urine, male children only).	Prevalence of *S. haematobium* is measured for the first time in Zanzibar.	Mansfield-Aders (1928)
1937–1938	Mozley describes the freshwater snails of Tanganyika and Zanzibar, and leads a study for snail control via environmental modification.	Perhaps the first successful elimination of *Bulinus africanus* species group through environmental modification (Mozley's ditch, Muyuni, Unguja).	Mozley (1939)
1962	Island wide parasitological and malacological surveys conducted on Unguja and Pemba.	Micro-epidemiological patterns of high prevalence demonstrated.	Goatly and Jordan (1965)

(*continued*)

Table 1 Milestones and research programmes contributing to schistosomiasis research and control on the Zanzibar islands. CCA = Circulating Cathodic Antigen, MDA = Mass Drug Administration, WHO = World Health Organization, ZEST = Zanzibar Elimination of Schistosomiasis Transmission. (*cont'd*)

Year	Milestone	Outcome	Reference (s)
1964–1967	Efficacy of newer chemotherapeutics assessed and delivery platform trialled in mass-treatment programmes directed at school and community.	Highlighted need for local authority and community cooperation in treatment programmes and that a drug of choice needs to be efficacious at low doses.	Forsyth (1966), Forsyth and Macdonald (1966), Forsyth and Rashid (1967b), Forsyth and Rashid (1967a), Macdonald et al. (1968b)
1975	World Health Organization (WHO) engagement is strengthened through visits from McCullough and Krafft investigating methods for the control of schistosomiasis in Zanzibar.	National plan for schistosomiasis control outlined, which includes chemotherapy, snail control and health education.	Mccullough and Krafft (1976)
1978	A syringe model for executing urine filtration is suggested for use in Zanzibar.	Leads to adoption of a more standardised and effective approach for performing urine filtration.	Mccullough (1978)
1978	Macrohaematuria is proposed as a proxy for *S. haematobium* infection for the first time in Zanzibar.	Allows for assessment of disease endemicity with few resources across Zanzibar.	Mccullough (1978)
1981	Urine filtration used to diagnose *S. haematobium* for the first time in Zanzibar.	Becomes the gold standard to which other diagnostic tests are compared.	Mgeni et al. (1990), Mgeni, (n. d.)

1984	WHO recommends praziquantel (Merck KGaA, Germany) for the treatment of schistosomiasis.	With an efficacious drug that works in a single dose, research priorities in Zanzibar move away from treatment methods.	World Health Organization (1985)
1986	Reagent strips to detect microhaematuria as a proxy for *S. haematobium* infection are used for the first time in Zanzibar.	A low-cost point of care diagnostic is found that serves as a good proxy for *S. haematobium* infection.	Savioli et al. (1989b), Savioli et al. (1989a), Savioli and Mott (1989), Lwambo et al. (1997b)
1986–1990	Test (haematuria) and treat (praziquantel) campaigns begin on Pemba.	Quicker proxy for determining infection status proves successful method for targeting schistosomiasis treatments in high prevalence areas, in turn reducing prevalence. Praziquantel proves efficacious form of treatment.	Savioli (1991a)
1988	Ivo de Carneri arrives on Pemba, sent by the Italian Cooperation to evaluate programmes initiated against schistosomiasis.	The importance of equipping Pemba with a public health laboratory is highlighted.	Confalonieri (2009)
1991	Success of WHO collaboration with the Government of Zanzibar in organising control of schistosomiasis recognised as a big achievement.	First considerations for implementing integrated control of other tropical diseases in Zanzibar and in other countries using the WHO technical support.	Barakamfitiye (1991), Savioli (1991b)

(*continued*)

Table 1 Milestones and research programmes contributing to schistosomiasis research and control on the Zanzibar islands. CCA = Circulating Cathodic Antigen, MDA = Mass Drug Administration, WHO = World Health Organization, ZEST = Zanzibar Elimination of Schistosomiasis Transmission. (*cont'd*)

Year	Milestone	Outcome	Reference (s)
1993	Soluble egg antigen enzyme-linked immunosorbent assay for the diagnosis of *S. haematobium* infection tested for the first time in Zanzibar.	The sensitivity of the test was high but the specificity was low.	Xue et al. (1993)
1994	National Helminth Control Programme established by Zanzibar MoH.	First mass drug administration scheme on Unguja and Pemba, pioneering large-scale preventive chemotherapy programme for co-administration of anthelmintic drugs.	Mohammed et al. (2008)
1994	Ivo de Carneri Foundation is set up with the main task in fundraising for resources to build public health laboratory.	Plans to build Public Health Laboratory begin, fundraising supported by town of Cles (Italy) in memory of Ivo de Carneri. WHO officers oversee plans and construction.	de Carneri (1994), Shauri (1994), Albonico (1997), Confalonieri (2009)
1994	Malacological surveys take place on Pemba and Unguja by Stothard and Rollinson.	Molecular species diagnostics first applied for differentiating *Bulinus* species present on the islands.	Stothard et al. (1997), Stothard and Rollinson (1997a)
2000	Public Health Laboratory - Ivo de Carneri is built in Chake Chake, Pemba.	Activity begins in many public health areas, including control of schistosomiasis.	Confalonieri (2009)

2000	*Bulinus globosus* confirmed as primary intermediate host on Unguja and Pemba, *B. nasutus* considered refractory.	Control efforts concentrated in regions only where *B. globosus* present.	Stothard et al. (2000)
2002	Helminth Control Laboratory Unguja set up by funds from The Health Foundation, United Kingdom.	Laboratory on Unguja now equipped with snail culture rooms.	Stothard et al. (2006b)
2003	President of Zanzibar launches the *Piga vita Kichocho* control programme on Unguja and Pemba.	Prevalence of schistosomiasis reduces further.	Stothard et al. (2006b)
2003	Urine-circulating cathodic antigen (CCA) strips to identify *S. haematobium* infections are tested for the first time in Zanzibar.	Low sensitivity of CCA proves they cannot be used for prevalence surveys.	Stothard et al. (2009c)
2003	Schistosomiasis Control Initiative begins managing the delivery of praziquantel as part of *Piga vita Kichocho*.	Streamlined MDA begins in Zanzibar.	Stothard et al. (2006b)
2006	Urine-albumin levels, urine's opacity (turbidity), and urinary tract pathologies are tested for the first time in Zanzibar.	Elevated urine-albumin excretion was associated with urinary tract pathologies, particularly lesions of the bladder wall, in adults.	Sousa-Figueiredo et al. (2009)
2009	First Chinese team sent to Zanzibar to carry out onsite control work.	Stimulated future collaborations.	Zhou et al. (2019)

(*continued*)

Table 1 Milestones and research programmes contributing to schistosomiasis research and control on the Zanzibar islands. CCA = Circulating Cathodic Antigen, MDA = Mass Drug Administration, WHO = World Health Organization, ZEST = Zanzibar Elimination of Schistosomiasis Transmission. (*cont'd*)

Year	Milestone	Outcome	Reference (s)
2011	Elimination of schistosomiasis targeted by the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) alliance.	Alliance of multiple stakeholders to deliver biannual treatments to both children and adults, and interventions aimed at interrupting *S. haematobium* transmission.	Knopp et al. (2012)
2013	UCAA2000 and UCAA250 assays to test for *S. haematobium* infection are conducted for the first time in Zanzibar.	High sensitivity of the test demonstrated, but current use limited due to the resources needed.	Knopp et al. (2015a)
2014	China, WHO and Zanzibar Government sign memorandum of understanding for cooperation on schistosomiasis control in Zanzibar.	Planning of future schistosomiasis control projects and establishment of laboratory on Pemba commences.	Yang (2021)
2016	*Schistosoma bovis* identified in focal regions on Pemba.	Complicates surveillance of *S. haematobium* in snail infections, risk to livestock health.	Pennance et al. (2016), Pennance et al. (2018)
2017	Final parasitological surveys during ZEST.	Urogenital schistosomiasis was eliminated as a public health problem from most areas in Zanzibar.	Knopp et al. (2019b), Knopp et al. (2019c)

2017	China-Zanzibar schistosomiasis control project launched on Pemba; trials China-made praziquantel and molluscicide formulations.	Both China-made praziquantel and molluscicide are effective, reduction in *S. haematobium* prevalence in defined regions.	Wang et al. (2019), Xing et al. (2021)
2019	*Bulinus nasutus* is found to be naturally infected with *S. haematobium* on Pemba.	Complicates surveillance of *S. haematobium* and broadens inferred geographic range for transmission.	Pennance et al. (2022a)
2020	A multidisciplinary intervention approach including active, reactive and passive surveillance is implemented in the north of Pemba.	Micro-targeting of interventions is explored. Hotspots receive a large-scale multidisciplinary intervention approach; low prevalence areas receive targeted surveillance-response after MDA is stopped.	Trippler et al. (2021b), Trippler et al. (2022b)
2021	Work begins on construction of three water dams in Kinyasini, Chaani (Unguja) and Pujini (Pemba).	Unknown consequences for *S. haematobium* transmission due to potential habitat expansion for *Bulinus* in these regions.	Personal observation, Fatma Kabole (October 2021)

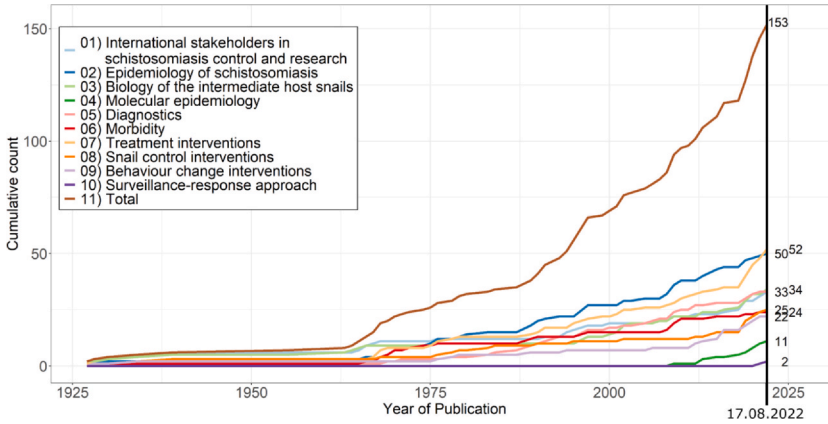


Fig. 2 Number of references included per section of the review and their year of publication based on literature search until 17th August 2022.

broadly stated that the parasite '*has been proved to be far commoner than previously thought*' (Mansfield-Aders, 1928). When the British colonialists realised that schistosomiasis was indeed highly prevalent and causing morbidity in the Zanzibari population, it became necessary to initiate island-specific research into the disease, and into the biology of the parasite and the snail host. Early research on Unguja by Mansfield-Aders (in his role as Economic Biologist for the Zanzibar Protectorate) and Cawston (an English physician and zoologist working in South Africa), identified transmission sites and the intermediate snail host species of the *Bulinus africanus* species group. In addition, a successful attempt was made to eliminate snail populations using environmental modification (Table 1).

Following the end of the Second World War, in- and out-patients for schistosomiasis were recorded in the Zanzibar Government hospitals between 1949 and 1951, although no further interventions to control the disease took place (Austin, 1955). During the 1960s, however, researchers from the East African Institute for Medical Research, the Ross Institute of Tropical Medicine and the London School of Hygiene and Tropical Medicine conducted multiple research studies investigating the associated morbidities for urogenital schistosomiasis in Zanzibar, and importantly testing diagnostics and measuring the efficacy of chemotherapeutic treatments, especially those delivered for the first time in mass-treatment programmes (Table 1). A decade after these pilot studies investigating mass-treatment programmes were performed, plans started to be drawn up between the Zanzibar government and WHO for a national control

programme for schistosomiasis. McCullough and Krafft, a WHO consultant and WHO Sanitary Engineer, respectively, spearheaded these plans, which saw the description of the first multifaceted control programme targeting the most endemic regions of Zanzibar (Table 1). This programme was designed to include simultaneous chemotherapy, snail control through molluscicide and health education (Table 1).

Although the plans were drawn up for national schistosomiasis control in the 1970s, it was not until 1986 that the first island-wide control programme was initiated on Pemba (Table 1). This island-wide test-and-treat campaign, which led to a reduction in *S. haematobium* prevalence and morbidity, was also the first to be conducted under international collaborative efforts. This test-and-treat programme was supported by the WHO and the Zanzibar MoH, and additionally by the Italian Agency for Development Cooperation and German Pharma Health (Savioli, 1991a). This engagement with Italian stakeholders was mediated by Savioli, an Italian physician residing on Pemba during this time who took on the role of Schistosomiasis Control Programme manager for Pemba (Table 1). Italian engagement led to a major development on Pemba through the construction of the Public Health Laboratory-Ivo de Carneri (PHL-IdC), first envisaged by Ivo de Carneri during a trip to Pemba in 1988, and then realised and completed posthumously in 2000 (Table 1). Unfortunately, Ivo de Carneri passed away before the total funds and approval to construct the laboratory were finalised (Shauri, 1994). Savioli and Albonico oversaw the building of the laboratory in their roles with the WHO, with a hope that the lab would serve as a '*Zanzibar Ministry of Health 'Station' for evaluating and developing strategies for control of major endemic diseases*' (Savioli, 1995).

In 1994, the pioneering National Helminth Control Programme on both Unguja and Pemba was initiated by the Zanzibar MoH, which marked the beginning of regular treatment for schistosomiasis using praziquantel (Table 1) (Mohammed et al., 2008). During this year, a long lasting collaboration was formed between the National Helminth Control Programme, Zanzibar and researchers from the Natural History Museum, London (NHM) whose priority was to identify the intermediate host snail species of the *B. africanus* species group responsible for transmitting *S. haematobium* on Unguja and Pemba (Table 1) (Stothard et al., 1997, Stothard and Rollinson, 1997a). The collaboration of the Zanzibar MoH with the NHM then led to MoH backing of the *Piga Vita Kichocho* campaign, which gained additional support from a partnership of donors including The Health Foundation (United Kingdom),

the African Development Bank and the recently founded Schistosomiasis Control Initiative (SCI, now Unlimit Health: www.unlimithealth.org).

From years of predominantly chemotherapy-based control efforts in Zanzibar, it became evident that MDA alone would not result in elimination. This led to the international Zanzibar Elimination of Schistosomiasis Transmission (ZEST) alliance in 2011 that had the primary objective to assist the Zanzibar government in its efforts to achieve elimination (Table 1) (Knopp et al., 2012). The alliance consisted of the Zanzibar MoH's NTD Programme, the PHL-IdC, the WHO, the SCI, the NHM, the Swiss TPH, and the Schistosomiasis Consortium for Operational Research and Evaluation (SCORE), University of Georgia, Athens, United States of America. SCORE funded operational research studies with the goal to provide pragmatic answers and to serve as proof-of-concept to inform control programmes committed to moving from morbidity control towards interruption of *Schistosoma* transmission. In Zanzibar, between 2011 and 2017, SCORE funded a large-scale cluster-randomised trial conducted in 90 randomly selected shehias to assess the impact of two intervention arms: biannual MDA + snail control (administering the molluscicide niclosamide to snail host infested freshwater) and biannual MDA + behaviour change interventions, on the prevalence and infection intensity of schistosomiasis in comparison to a third intervention arm of biannual MDA alone.

During the ZEST trial, and respective interventions, schistosomiasis was eliminated as a public health problem from most areas in Zanzibar, however, transmission of *S. haematobium* was not completely interrupted (Knopp et al., 2019b, Knopp et al., 2019c). Considerable spatial heterogeneity of *S. haematobium* infection was observed, including many low transmission shehias and a few shehias with persistent schistosomiasis transmission hotspots (Pennance et al., 2016). The challenge of spatial heterogeneity is being addressed as part of the ongoing SchistoBreak study led by researchers from the PHL-IdC and the Swiss TPH (Trippler et al., 2021b). In the SchistoBreak study, a multidisciplinary intervention approach is being implemented in high transmission shehias (>2% and/or >3% *S. haematobium* prevalence in community-based or school-based surveys, respectively), consisting of MDA, snail control and behaviour change interventions. In shehias with a low prevalence of *S. haematobium*, a surveillance-response approach with active (school-based test-and-treat), reactive (test-and-treat in households of positive-tested schoolchildren, and

test-and-treat and focal snail control at identified transmission sites) and passive (test-and-treat in health facilities for patients presenting with *S. haematobium* infection symptoms) surveillance is being implemented instead of MDA (see section Implementation of surveillance-response interventions in Zanzibar). The study interventions aim to trigger an appropriate response in the high transmission shehias, and to prevent resurgence in the low transmission areas, without over-treating a mostly healthy population. A China-Zanzibar schistosomiasis control project conducted in four shehias in Pemba (Tangani, Wingwi, Kiyuyu and Uwandani), with three additional shehias monitored as control groups (Wambaa, Vitongoji and Shengejuu) (Yang et al., 2021), coincided with the end of ZEST interventions, also with an overarching goal to help reach schistosomiasis elimination in Zanzibar, while testing the efficacy of China-made praziquantel and molluscicides, respectively (Table 1).

5. Epidemiology of schistosomiasis in Zanzibar

5.1 *Schistosoma* spp. on Unguja and Pemba

Unlike urogenital schistosome transmission, autochthonous transmission of intestinal schistosomes does not occur in Zanzibar, mainly because *Biomphalaria*, a genus containing keystone intermediate snail host species for *Schistosoma mansoni*, is absent from the islands. However, two studies, one in 1964 and one in 1965, have noted, that three children, who had not travelled off the islands, passed apparent *S. mansoni* eggs in their faeces (Forsyth and Macdonald, 1966, Forsyth, 1969). No other inference of locally transmitted *S. mansoni* infections on Unguja and Pemba have ever been reported. Subsequent parasitological surveys suggest that the only other occurrences of *S. mansoni* infections reported from Zanzibari's were imported infections, acquired elsewhere (Mccullough & Krafft, 1976, WHO Archives, n.d.). In addition, malacological surveys across Unguja and Pemba have not found any snails of the genus *Biomphalaria* (see section Biology of the intermediate host snails). Therefore, it is possible that these earlier reports of *S. mansoni* infections were due to incorrect recollections (or recordings) of travel histories and/or misidentification of parasite eggs in the faeces (see section Diagnostics). As detailed in the following sections, *S. haematobium* and *S. bovis* are currently the only endemic *Schistosoma* species on the islands and prior to 2016 *S. haematobium* was the only endemic species recorded.

5.2 *Schistosoma haematobium* prevalence changes over 100 years

As shown in Fig. 3, the earliest prevalence of *S. haematobium* infection was recorded as 11.9% on Unguja in 1927 and as 58.6% on Pemba (males only) in 1962 (Mansfield-Aders, 1928, Goatly and Jordan, 1965). The majority of early studies that tested for *S. haematobium* infections on the Zanzibar islands used urine analysis and egg-detection, and were conducted on Unguja, particularly during the 1960s and 1970s. Across these two decades, the aim for many studies was to determine efficacious treatment approaches to reduce morbidity in individuals attending specific schools or residing in particular communities (see sections Morbidity, and Treatment interventions) (Macdonald et al., 1968b, Mgeni et al., 1990).

These surveys showed the heterogeneous nature of *S. haematobium* transmission; prevalence varied at different locations at the same time point. For example, on Unguja in 1966, the *S. haematobium* prevalence reported at Kinyasini school was 87.6% yet 22.4% was reported at Donge school (Fig. 3) (Macdonald et al., 1968a, Macdonald et al., 1968b). Over a decade later in 1979, the overall prevalence in Donge schoolchildren was recorded at 27.0%, showing that transmission in this region had remained consistent over this time period (Muhammed, 1980). During the 20th century, the majority of parasitological surveys measuring the prevalence of *S. haematobium* on Unguja and Pemba were from small-scale surveys in selected schools or shehias. One survey that is detailed in a WHO report serves as an exception to this, where a prevalence of 14.5% was recorded based on a survey conducted with 9,473 participants from 61 different schools on Unguja in 1976, and thus, represents the pioneer of large-scale studies concerning *S. haematobium* prevalence on the Zanzibar islands (WHO Archives, n.d.).

The 1980s began with parasitological surveys done in Kinyasini, Unguja, which showed a focal prevalence of 49.3% in 1981, 39.8% in 1982 and 28.92% in 1983 (Mott et al., 1983, Mgeni et al., 1990). These surveys were also the first to measure heavy-intensity infection (defined as ≥50 eggs in 10 mL of filtered urine) in Zanzibar, with the highest heavy-intensity infections in children aged 10–14 years (30.2%) and no significant difference between girls and boys (Mgeni et al., 1990). In general, however, in the 1980s and 1990s, the research focus, and therefore records of *S. haematobium* prevalence, switched from Unguja to Pemba, with no studies of *S. haematobium* prevalence between the years of 1984–2001 on

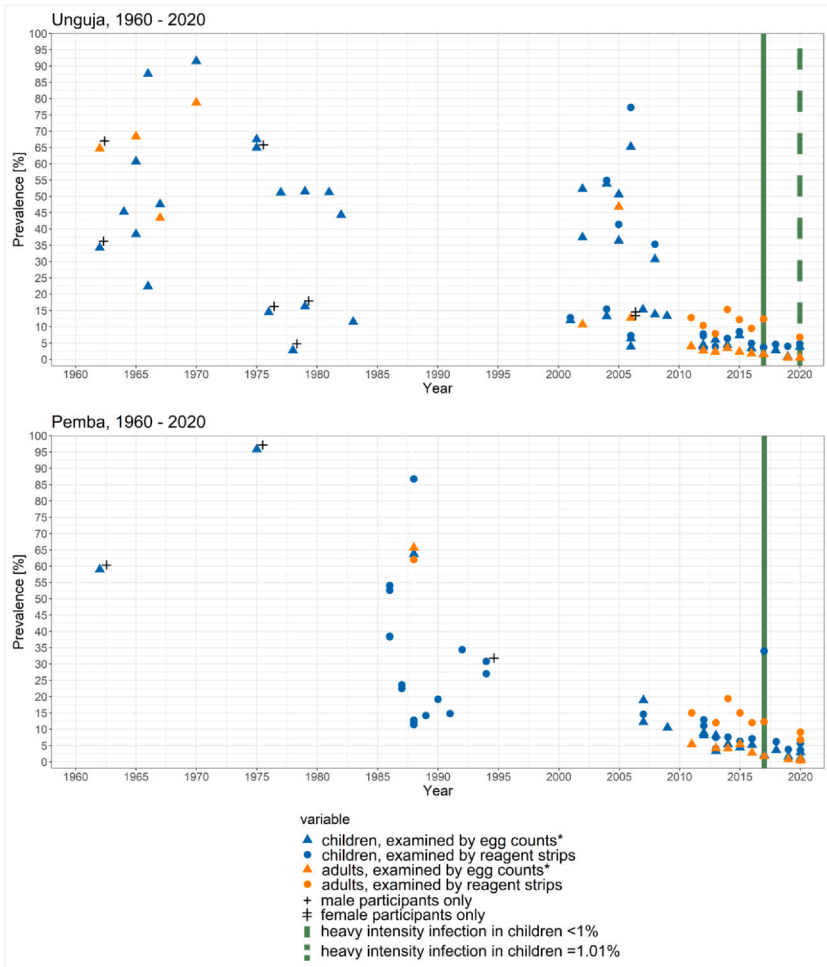


Fig. 3 *Schistosoma haematobium* prevalence in children and adult populations detected by egg counts (*by urine sedimentation until 1979, by urine filtration since 1980) or reagent strips on Unguja and Pemba between the years 1960–2020 reported in 38 records (Goatly and Jordan, 1965, Forsyth and Macdonald, 1966, Macdonald et al., 1968a, Macdonald et al., 1968b, Forsyth, 1969, Forsyth, 1971, Manning, 1976, Mccullough & Krafft, 1976, Haji and Mgeni, 1980, Savioli et al., 1989a, Savioli et al., 1989b, Savioli and Mott, 1989, Mgeni et al., 1990, Savioli et al., 1990b, Savioli, 1991a, Alawy, 1993, Albonico et al., 1997, Lwambo et al., 1997b, Stoltzfus et al., 1997b, Mgeni, 2002, Stothard et al., 2002d, Rollinson et al., 2005, Rudge et al., 2008, Sousa-Figueiredo et al., 2008, Sousa-Figueiredo et al., 2009, Stothard et al., 2009a, Stothard et al., 2009b, Stothard et al., 2009c, Guidi et al., 2010, Knopp et al., 2010, Knopp et al., 2013a, Knopp et al., 2013b, Knopp et al., 2015b, Knopp et al., 2019b, Kim et al., 2020, Trippler et al., 2021a, Trippler et al., 2022b, Mgeni, n. d.). One point represents the *S. haematobium* or microhaematuria mean prevalence per publication even if prevalence in publications

(Continued)

Unguja (Fig. 3). The main diagnostic method during these decades on Pemba was reagent strips to test for microhaematuria (blood in urine) (the highest reported microhaematuria prevalence was 86.8% in one school in 1988) (Fig. 3) (Savioli et al., 1990b).

During the 2000s, there was a recommenced focus on Unguja, as evidenced by several studies (Fig. 3). The first study in the decade was conducted in 10 schools in Unguja and aimed to investigate the correlation between *S. haematobium* and the distribution of *B. globosus* and *B. nasutus* and showed a mean *S. haematobium* prevalence of 12% (Fig. 3) (Stothard et al., 2002d). The 2000s also saw the highest ever recorded prevalence of *S. haematobium* in school-aged children (boys and girls) on both Unguja and Pemba. Diagnosis was based on urine filtration implemented in multiple schools. On Unguja in 2008, this prevalence was 30.7% in children from five different schools (Stothard et al., 2009c). For Pemba in 2007, the highest comparable prevalence was reported as 18.9% in children from eight different schools (Guidi et al., 2010). A few years later, in 2011, annual large-scale surveys started for the first time in schools and communities on both islands and with urine filtration and reagent strips used to determine prevalence. The surveys showed a prevalence based on urine filtration of 4.3% and 2.7% in Unguja and of 8.9% and 5.5% in Pemba in schoolchildren and adults, respectively (Knopp et al., 2012, Knopp et al., 2013a). Microhaematuria was detected in 7.3% and 10.4% in Unguja and in 11.1% and 14.3% in Pemba schoolchildren and adults, respectively (Knopp et al., 2013a). This was the beginning of the ZEST project, a cluster-randomised trial that was implemented from 2012 to 2017 in communities and schools within 90 shehias on Unguja and Pemba. The ZEST project included multidisciplinary interventions (see sections Treatment interventions, Snail control interventions and Behaviour change interventions) and annual parasitological surveys (Knopp et al., 2019b). These annual monitoring surveys continued beyond ZEST and until 2020

Fig. 3—Cont'd is reported separately per schools/shehias. The green solid line indicates the year where the islands reached elimination as a public health problem according to the WHO definition provided in the Neglected Tropical Diseases roadmap 2020–2030 (World Health Organization, 2020). The green dashed line represents the year Unguja rebounded to above 1% prevalence. A prevalence of 11.9% in boys on Unguja in 1927, reported by Mansfield-Aders (1928) and a prevalence of 38% and 31.2% based on urine sedimentation in participants aged 5–15 years and aged 16–55 years, respectively, reported by Mccarthy (1930), was excluded from the figure to improve the visualisation of the majority of data points.

(Trippler et al., 2021a). Towards the end of this decade of intense efforts for reaching elimination, Unguja and Pemba reached their all-time lows of island-wide *S. haematobium* prevalence in school-aged children: Unguja with 0.8% in 2019 and Pemba with 1.2% in 2020 (Trippler et al., 2021a, Trippler et al., 2022b). This lowest prevalence of 1.2% for Pemba is also the most recent prevalence data published, while the most recent prevalence reported for Unguja is 3.4% in 2020 (Trippler et al., 2021a). Heavy-intensity infections occurred in 0.6% and 2.9% of school-aged children on Unguja and Pemba, respectively, at the start of ZEST in 2012, and had decreased to 0.1% and 0.6%, respectively, in 2019 (Knopp et al., 2019b, Trippler et al., 2021a). However, heavy-intensity infection then increased again to 1.0% and 0.7%, respectively, in 2020 (Trippler et al., 2021a).

5.3 Spatial heterogeneity of *Schistosoma haematobium* infections in Zanzibar

As alluded to above, a detailed examination of the results reveals that a spatial heterogeneity of *S. haematobium* prevalence exists across Unguja and Pemba, a pattern already striking in 1962 when on Unguja 89.0% of the population of the village Mtende were infected with *S. haematobium* while 0% of the surveyed populations of Ungujaukuu and Ndijani were found to be positive (Goatly and Jordan, 1965). The heterogeneity on Pemba was similarly pronounced within this same study: in Kengeja, 96.0% of the surveyed individuals tested positive for *S. haematobium* while only 20.0% of the participants in Konde were infected (Goatly and Jordan, 1965). Spatial heterogeneity of *S. haematobium* within the Zanzibar islands has been a theme in many other studies throughout the history of epidemiological research in Zanzibar, which have provided several important findings (Chopra, 1968, Stothard et al., 2002d, Rollinson et al., 2005, Rudge et al., 2008, Sousa-Figueiredo et al., 2009, Stothard et al., 2009a, Stothard et al., 2009c, Knopp et al., 2010, Ali et al., 2015, Pennance et al., 2016, Knopp et al., 2019b, Trippler et al., 2021a, Trippler et al., 2022b). A study conducted around the school of Donge on Unguja from 1966 to 1968, revealed that the spatial heterogeneity of *S. haematobium* infections even occurs on a very small-scale level within shehias (Forsyth and Macdonald, 1966, Macdonald and Forsyth, 1968, Macdonald et al., 1968a). Many years later, in 2005, 2013 and 2020/2021, the proximity of homes, schools and shehias to water bodies (with *B. globosus*) was reported as a risk factor for *S. haematobium* infection on Unguja and Pemba (Rudge et al., 2008, Pennance et al., 2016, Trippler et al., 2021a, Trippler et al., 2022b). These

results were key to understanding the mode of *S. haematobium* transmission on the islands and provided crucial information on how to determine, and subsequently where to implement, targeted interventions in Zanzibar on the last mile towards schistosomiasis elimination.

Due to the spatial heterogeneity of *S. haematobium* transmission, shehias with a persisting high prevalence of *S. haematobium* prevented the islands from reaching the aim 'elimination as a public health problem' according to the definition provided in the WHO progress report 2001–2011 and strategic plan 2012–2020, which stated that each sentinel site (which in the case of Zanzibar we define as each shehia) should have a *S. haematobium* prevalence <1% (World Health Organization, 2013). Since throughout the years there was at minimum one sentinel site with a *S. haematobium* prevalence ≥1% on each island (Knopp et al., 2019b), none of the islands would yet technically aim for interruption of transmission by definition. However, the definition for 'elimination as a public health problem' was refined in the roadmap for NTDs 2021–2030 'as <1% proportion of heavy intensity schistosomiasis infections', i.e. removing 'in all sentinel sites' from the goal (World Health Organization, 2020). According to this new definition and the *S. haematobium* prevalence calculated in Knopp et al. (2019b) and Trippler et al. (2021a), Unguja reached 'elimination as a public health problem' by 2018, although rebounding to slightly above 1% in 2020, whilst Pemba reached the same goal in 2017, and has been aiming for interruption of transmission ever since (Fig. 3).

5.4 Sex-related difference in *Schistosoma haematobium* prevalence

Typically, higher *S. haematobium* prevalence is observed in boys than in girls in Zanzibar, mentioned first in a study conducted in 1966 (Forsyth and Macdonald, 1966). Many publications followed that reported this (statistically significant) sex-related difference (reported Odds Ratios are shown in Fig. 4) (Forsyth and Macdonald, 1966, Forsyth, 1969, Savioli et al., 1989b, Albonico et al., 1997, Stoltzfus et al., 1997a, Rudge et al., 2008, Sousa-Figueiredo et al., 2009, Stothard et al., 2009a, Knopp et al., 2013a, Kim et al., 2020, Trippler et al., 2021a). A sex disparity was also reported in many *S. haematobium* studies completed in other sub-Saharan African countries, such as Nigeria or Zambia (Agnew-Blais et al., 2009, Atalabi et al., 2016, Tembo et al., 2022). The shared explanation is that boys have a higher exposure to open freshwater bodies (Albonico et al., 1997, Rudge et al., 2008, Agnew-Blais et al., 2009, Atalabi et al., 2016, Tembo et al., 2022)

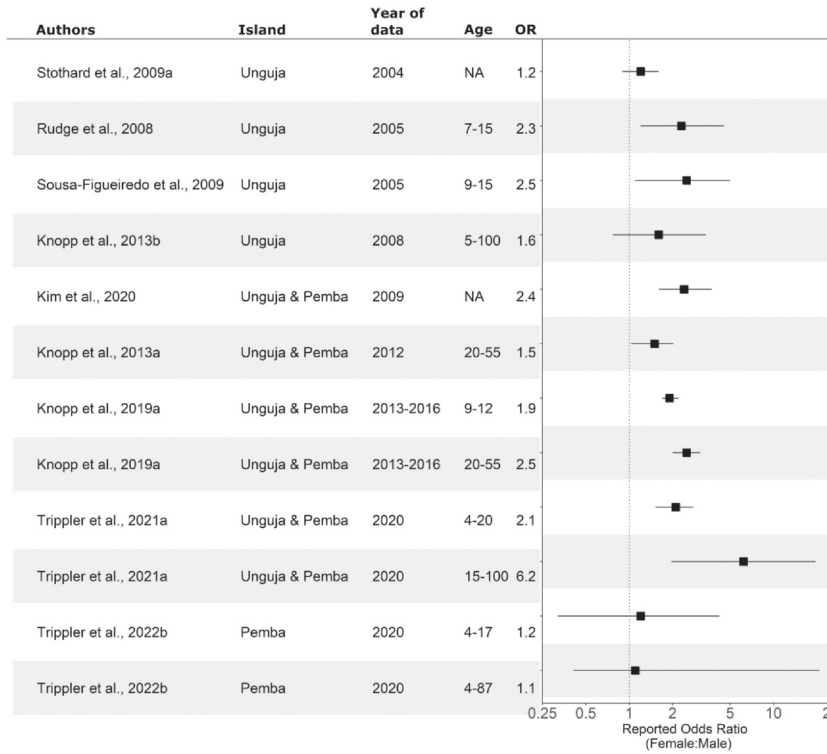


Fig. 4 Sex-related (Female:Male) Odds Ratios (ORs) and 95% confidence intervals for *Schistosoma haematobium* infections on Unguja and Pemba in the years 2004–2020, reported by nine publications, and sorted by the year when the data were collected (Rudge et al., 2008, Sousa-Figueiredo et al., 2009, Stothard et al., 2009a, Knopp et al., 2013a, Knopp et al., 2013b, Knopp et al., 2019a, Kim et al., 2020, Trippler et al., 2021a, Trippler et al., 2022b). Points to the right of the dashed line indicate that the chance of being infected with *S. haematobium* is higher in males than in females.

because of, for example, pastimes including fishing and swimming, which in these countries are activities more commonly done by males (Knopp et al., 2013a, Atalabi et al., 2016, Tembo et al., 2022). Furthermore, in Zanzibar, girls often take part in separate activities than boys from the age of 6 or 7 onwards due to their Muslim culture, and young adolescent girls are more self-conscious about bathing in local water bodies (Person et al., 2016a). Some studies also reported a (statistically significant) higher prevalence in male adults compared with female adults in Zanzibar (Forsyth, 1969, Rudge et al., 2008, Knopp et al., 2013a, Trippler et al., 2021a, Trippler et al., 2022b), although one publication also reported a higher prevalence in female than in male study

participants (Savioli, 1991a). The most recent publication, which included results from both islands showed that in 2019, even with a very low overall prevalence of 1.2%, male study participants <40 years of age showed a higher prevalence than female study participants <40 years of age, particularly in the age group of 8–16 years (Trippler et al., 2021a). A treatment gap of 16 months, followed by a resurgence of the overall prevalence, made the sex difference even more apparent since a higher proportion of males aged <60 years was infected than females aged <60 years, which was particularly evident in the age group of 8–20 years (Trippler et al., 2021a).

5.5 Age-related difference in *Schistosoma haematobium* prevalence

Besides the difference in *S. haematobium* infection between males and females, many studies have also tested for variances of *S. haematobium* prevalence between age groups in Zanzibar. However, most of these studies differ in the grouping of ages and while some of the results are based on data that were collected before treatment, others are based on data after several rounds of treatment, which might make a significant difference as shown in Trippler et al. (2021a). Therefore, it is not possible to clearly compare the results of the different studies with each other. Despite these limitations for comparison, there are interesting patterns in findings such as from Macdonald et al. (1968a), who reported a statistically significant difference between age groups in one of the first studies in Zanzibar with a higher *S. haematobium* prevalence in children aged ≤13 (40.2%) than in children aged ≥14 (26.5%) in specific regions around Donge school on Unguja. In contrast, many other studies conducted in Zanzibar showed that the prevalence was highest in other age groups such as children aged >12 years (Stothard et al., 2009a), 12–14 years (Knopp et al., 2010), 13–17 years (Trippler et al., 2022b), or 15–19 years (Savioli et al., 1990b). In general, studies have shown that children and adolescents tend to have the highest prevalence and are therefore most in need of interventions against schistosomiasis and associated morbidity. While many studies have been focussing on school-aged children, the ZEST study also specifically assessed prevalence data for first year students in 2011/2012 prior to the onset of the study (5.2% in Unguja and 12.2% in Pemba) and again in 2017 (2.3% in Unguja and 2.8% in Pemba), finding that a higher *S. haematobium* prevalence was recorded at both time points in this age group on both islands than in other age groups (9–12 years: 4.1% in Unguja and 8.2% in Pemba in 2011, 1.7% in Unguja and 1.7% in Pemba in 2017; adults

>55 years: 2.5% in Unguja and 5.5% in Pemba in 2011, 1.3% in Unguja and 1.7% in Pemba in 2017), and a higher proportion with heavy intensity infections (Knopp et al., 2019b).

5.6 Outlook

The tremendous decrease of *S. haematobium* infections over the past 100 years on Unguja and Pemba, exemplified by the reduction in *S. haematobium* prevalence from >50% historically down to <5% in 2020, is undoubtedly and unequivocally evident. However, a closer look reveals a striking geospatial heterogeneity of *S. haematobium* transmission on both islands of Zanzibar and a disparity in infection incidence between sexes, boys more often infected than girls in most of the studies reported. It is hoped that if the islands remain on the declining trajectory of *S. haematobium* prevalence and continue to target interventions appropriately, elimination as public health problem in all sentinel sites as well as interruption of transmission, with zero incidences of schistosomiasis cases, could be a realistic outcome within the near future.

6. Biology of the intermediate host snails

6.1 Endemic *Bulinus* species of Unguja and Pemba

Using the currently accepted taxonomic organisation of the *Bulinus* genus given by Brown (1994) with the addition of one currently unidentified species (discriminated by molecular DNA markers), five endemic *Bulinus* species are known across Unguja and Pemba. These five species include *B. globosus* and *B. nasutus* of the *Bulinus africanus* species group, in addition to *B. forskalii* and two other cryptic taxa of the *Bulinus forskalii* species group that are conchologically indistinguishable from *B. forskalii*, namely *B. barthi* and an undetermined species provisionally referred to as *B. forskalii* sp. (Stothard et al., 2002b, Kane et al., 2008, Stothard et al., 2013). As discussed in detail below, *B. globosus* is recognised as the primary intermediate host for *S. haematobium* on both islands (Stothard and Rollinson, 1997b) and, more recently, for the cattle parasite *S. bovis* on Pemba (Pennance et al., 2018). However, *B. nasutus* may also act as an intermediate host for *S. haematobium* in specific foci under certain circumstances (Pennance et al., 2022a). The primary focus of the following section is to provide an overview of the involvement, identification, distribution and genetic diversity of *B. globosus* and *B. nasutus* on Unguja and Pemba. Despite years of malacological surveillance, the absence of *Biomphalaria*

species on both Unguja and Pemba, perhaps due to the higher surface temperatures on the islands as in coastal plains of Tanzania and Kenya that inhibit colonisation of this genus (Sturrock, 1965, Sturrock, 1966), diminishes any concerns for the introduction and transmission of *S. mansoni* here.

Historically, the endemic species of the *B. forskalii* species group have been less studied than the *B. africanus* species group, since none have been of concern for local transmission of *Schistosoma* on Unguja and Pemba or neighbouring Mafia Island (Tanzania), and experimental infections have shown a refractory nature to schistosome infection (Stothard et al., 2002b, Stothard et al., 2013). Datasets from three recent studies recorded *B. forskalii* species group occurrence across the islands, with the exception of the southern region of Unguja (Fig. 5) (Pennance et al., 2016, Allan et al., 2020, Pennance, 2020). Cohabitation of *B. forskalii* species group with *B. africanus* species group is commonly observed in water bodies on both Unguja and Pemba (Mozley, 1939, Stothard et al., 1997, Pennance et al., 2016). Although the presence of *B. forskalii* group snails was established on the Zanzibar islands in the 1930s (Mozley, 1939), the two morphologically cryptic taxa were delimited nearly 70 years later upon starch gel electrophoresis of isoenzymes in the case of *B. barthi*, and DNA sequencing of the nuclear internal transcribed spacer (ITS) of the ribosomal gene complex and mitochondrial cytochrome oxidase subunit 1 (*cox*1) gene in the case of *B. forskalii* sp. (Stothard et al., 2002b, Kane et al., 2008, Stothard et al., 2013). From the sparse data available, *B. barthi* and *B. forskalii* sp. appear to have a limited distribution across Unguja and Pemba, being identified from only one and three locations, respectively (Fig. 5). Perhaps further locations for these cryptic species, as well as the distribution of *B. forskalii* relative to these cryptic taxa, might be discovered with increased sampling combined with molecular species identifications. The genetic divergence of Zanzibari *B. barthi* and the *B. forskalii* sp. relative to other endemic Indian Ocean island species such as *B. cernicus* and *B. bavayi*, has been shown in a number of studies (Stothard et al., 2002b, Kane et al., 2008, Jørgensen et al., 2011). Whether the islands' *B. forskalii* group snails are naturally penetrated by *S. haematobium* group miracidia is yet to be established, however this could be an interesting tool to include in transmission surveillance using molecular xenomonitoring.

6.2 Taxonomic history and identification of *Bulinus globosus* and *Bulinus nasutus*

In 1926, Cawston along with Mansfield-Aders collected snails from an ornamental lily pond, constructed of cement, within the walls of the

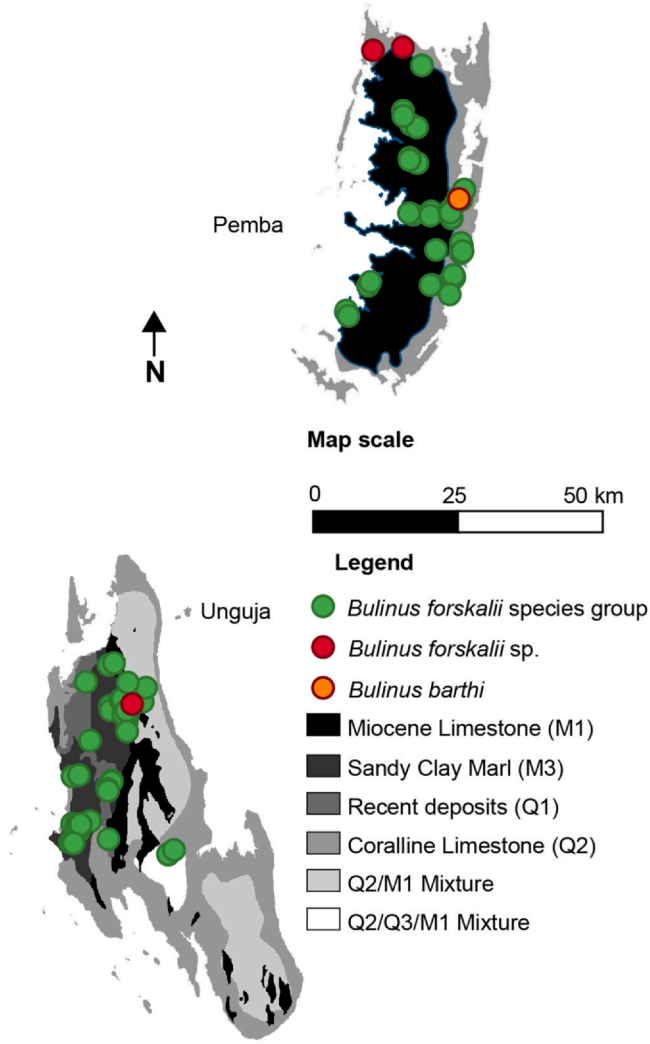


Fig. 5 Distribution of *Bulinus forskalii* species group snails on Unguja and Pemba, United Republic of Tanzania, determined morphologically as the *Bulinus forskalii* species group according to Brown (1994) (green), or by molecular markers to species level for isolates identified as the currently undescribed *Bulinus forskalii* sp. (red) and *Bulinus barthi* (orange) identified in Stothard et al. (2002b) and Kane et al. (2008). Distribution data for species of the *B. forskalii* species group presented here were taken from presence/absence reported from malacological surveys (Supplementary File 2: Table 2) conducted on Unguja and Pemba from 2011 to 2018 (Pennance et al., 2016, Allan et al., 2020, Pennance, 2020). The base map represents known geological features of Unguja and Pemba (United Nations, 1987, Hardy et al., 2015).

Maruhubi Palace ruins (approximately 4 km north of Stone Town, Unguja) and identified them as *Isidora (Physopsis) ovoidea* based on Bourguignat's (1879) description of *Physopsis ovoidea* from Bagamoyo, coastal Tanzania (Cawston, 1927b, Mansfield-Aders, 1928). Thirty years after Cawston's and Mansfield-Aders' collections, the species *I. ovoidea* was collapsed into the species *Bulinus africanus* (Mandahl-Barth, 1957, Brown, 1994). Being familiar with identifying *I. africana* (*B. africanus*) in South Africa (Cawston, 1924), Cawston (1927b) noted that these *I. ovoidea* from Maruhubi were 'distinct from *Isidora* (*Physopsis*) *africana* (Krauss) and not a variety of the same species'. However, snails from a second locality, a highly endemic region neighbouring the Mwera river (~7 km east of Maruhubi), were notably much harder to distinguish morphologically from South African *Physopsis africana*, with an intuitive comment that it 'is at times a similar difficulty in distinguishing the shells of *Isidora* (*globose*)' (Cawston, 1927b). It has been established that snails from the Maruhubi ponds and the Mwera river region are *B. globosus* (Rollinson, unpublished observations). Observations by Cawston therefore highlight the morphological plasticity of the *B. africanus* species group on Unguja (Fig. 6).

Further attention was given to the freshwater snail fauna of Unguja and Pemba between 1937 and 1938, including notes on habitat preferences and photographing snails, showing the considerable morphological polymorphism that existed (Mozley, 1939). Despite the morphological plasticity of the *Bulinus* collected, Mozley (1939) determined that only one species, which he referred to as *Physopsis globose* (Morelet), was present across the Zanzibar islands.

Although, the next malacological surveys were not conducted until 1962, it is of note that great efforts were made between the years 1955 and 1965 to describe and organise species of the genus *Bulinus* by Mandahl-Barth (WHO Archives, 1955). The work supported by the WHO first resulted in a revised snail taxonomy published with refinements in 1958 (Mandahl-Barth, 1958). Following this, Mandahl-Barth requested, and received funding from donors and the WHO to expand his work in setting up a self-governing institution for intermediate host snail identification, the Danish Bilharziasis Laboratory (DBL), in Copenhagen, Denmark (Mandahl-Barth, 1963). Following malacological surveys in September 1962, just prior to the formal establishment of the DBL in July 1964 (Candau, 1964), 41 snails from different locations in Zanzibar were sent to Mandahl-Barth for identification (Mandahl-Barth, 1963, Goatly and Jordan, 1965). All the snails were identified by Mandahl-Barth as

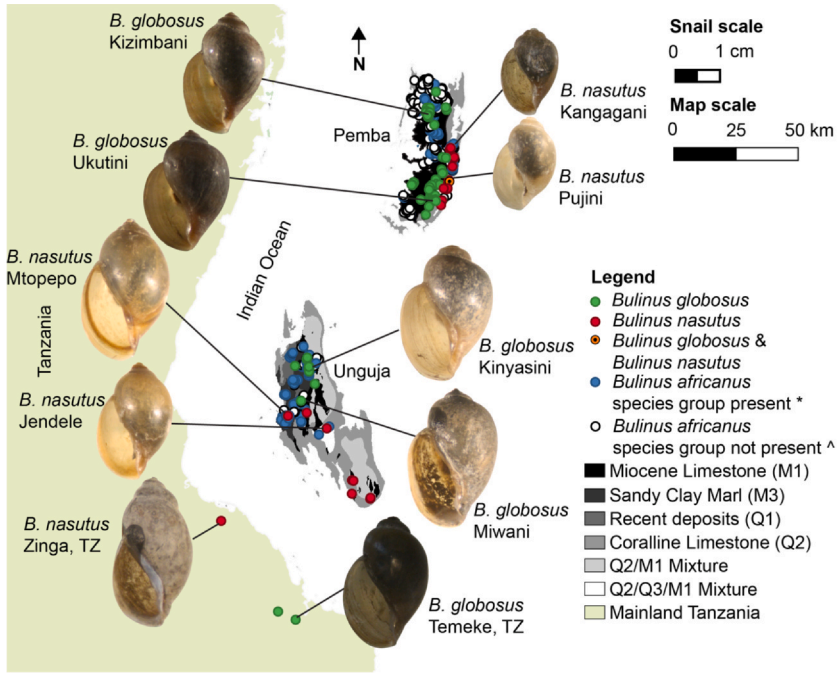


Fig. 6 Distribution of *Bulinus globosus* and *B. nasutus* on Unguja, Pemba, and coastal Tanzania, as summarised by available presence/absence data with geocoordinates from previous studies (Stothard et al., 1997, Kane et al., 2008, Pennance et al., 2016, Knopp et al., 2019c, Allan et al., 2020, Pennance, 2020, Pennance et al., 2022a, Trippler et al., 2022b) (Supplementary File 2: Table 2). Where DNA sequence data was available, this was used to distinguish *B. globosus* and *B. nasutus*. Selected photographs of identified species shells are shown to demonstrate the morphological plasticity of both *B. globosus* and *B. nasutus* across Unguja, Pemba and coastal Tanzania (Pennance, 2020). The base map represents known geological features of Unguja and Pemba (United Nations, 1987, Hardy et al., 2015). *Represent snails identified morphologically, and therefore only to *B. africanus* species group level. ^Represents specific freshwater sites on Unguja and Pemba where *B. africanus* species group snails have not been found (absent).

B. nasutus, except for snails collected from two sites in northern Unguja (neighbouring Donge and Chaani) that were considered *B. globosus* (Goatly and Jordan, 1965), thus creating a malacological map at clear odds with Mozley's (Mozley, 1939).

Throughout the 1970s and 1980s, enzyme data on *Bulinus* species were collected, including enzymatic markers for *B. nasutus* and *B. globosus* from the coastal region of Tanzania (Rollinson and Southgate, 1979). Such

enzymatic profiling, by isoelectric focusing in polyacrylamide gels, contributed significantly towards a revised, and currently mostly agreed upon, taxonomy for the genus *Bulinus* (Brown, 1994). Taxonomic boundaries between the 12 nominal species placed within the *B. africanus* species group, including those from Zanzibar, still remained unclear (Brown, 1995). Work on enzymatic markers for *Bulinus* proved regionally specific (Stothard et al., 1997, Stothard and Rollinson, 1997a). Therefore, after the development of specific *B. globosus/nasutus* enzymatic markers for Unguja and Pemba, an updated malacological map created by molecular identifications was produced, showing the allopatric distribution of *B. globosus* and *B. nasutus* (Stothard et al., 1997). Analysis of multiple shell measurements relative to the species inferences made by enzymatic markers, revealed that *B. globosus* and *B. nasutus* were impossible to differentiate by conchological features (Stothard et al., 1997). It was recommended thereafter to refer only to *B. globosus* and *B. nasutus* to the species level when molecular identification had been employed on the specimens in hand, or samples from the same locality have had prior identification to allow for some inferences of species based on the locality.

From 1990 onwards, with increasing access to PCR and DNA sequencing technologies, several rapid diagnostic assays to differentiate *B. globosus* and *B. nasutus* on the Zanzibar islands were developed, including a random amplified polymorphic DNA (RAPD) assay (Stothard et al., 1997), two diagnostic restriction fragment length polymorphism (RFLP) (Stothard and Rollinson, 1997b, Stothard and Rollinson, 1997a) and a multiplex SNaPshot™ assay (Stothard et al., 2002a). Although RAPD, RFLP and the SNaPshot™ assay still have their importance for cheap and quick distinction between *B. globosus* and *B. nasutus*, all have become somewhat redundant owing to the affordability of DNA sequencing. Producing DNA barcodes for collected *Bulinus*, usually the *cox*1 and nuclear ITS regions, and inferring species based on comparisons to reference sequences for *Bulinus* through phylogenetic analysis is an excellent method for molecular identification and characterisation for these taxa (Rollinson et al., 2009a). Thanks to the work of Kane et al. (2008), a comprehensive range of primers are available to perform PCR and sequencing on all major groups of *Bulinus* to target barcoding regions, including those from Zanzibar (Kane et al., 2008). Using *cox*1 barcoding, recent identification of *Bulinus* snails collected across Unguja and Pemba, complemented the distribution data that was reported from the last major mapping exercise (Stothard et al., 1997), with the minor exception of a few

localities where *B. globosus* and *B. nasutus* cohabit (Fig. 6) or are close neighbours (Pennance et al., 2022a). Although the snails on Unguja and Pemba are referred to throughout this review as *B. globosus*, it is clear from the comparative mitogenomic data currently available that multiple disparate lineages from Unguja, Pemba and mainland Africa represent a *B. globosus* species-complex (Pennance, 2020, Zhang et al., 2022). Classification of *B. globosus* as either a widely distributed highly variable species or split into formally named species/sub-species is to be established with wider sampling and analysis on the species boundaries.

6.3 Incrimination of *Bulinus* as intermediate host(s) of *Schistosoma haematobium*

Bulinus sp. was first incriminated as an intermediate host on Unguja by Mansfield-Aders who observed *Schistosoma* cercariae from nine (3.6%) dissections of 249 *B. africanus* species group snails found in 'ponds and swamps from various parts of Unguja [*sic*]' (Mansfield-Aders, 1928, Mccarthy, 1930) subsequently identified schistosome cercariae from seven (1.8%) dissections of 390 *B. africanus* species group snails from Wete [*sic*], Pemba. Further work was proposed to continue during the early 1930s for identifying the vector snails in Zanzibar (Zanzibar Protectorate, 1930–1934), possibly because of the uncertainty in the snail species identification and the absence of patent (i.e. shedding) schistosome infections from collected snails during these early collections.

From the late 1990s, with the capacity to accurately identify *Bulinus* specimens to the species level, it became possible to explore the compatibility of *B. globosus* and *B. nasutus* to different isolates of *S. haematobium*. During experimental infection studies, *B. globosus* from Unguja were shown to be susceptible to isolates of *S. haematobium* originating from both Unguja and Pemba, whereas *B. globosus* from Pemba were only susceptible to the *S. haematobium* isolated from Unguja (Table 2), even when repeated attempts to infect the same Pemba *B. globosus* individuals with Pemba *S. haematobium* were performed (Stothard and Rollinson, 1997b). Despite the results from this small experimental study (Table 2), it is certain that transmission of *S. haematobium* on Pemba is mostly autochthonous, and not only imported *S. haematobium* from Unguja as these results may suggest. It is hence suspected that another factor was the cause of the unexpected resistance of Pemba *B. globosus* to Pemba *S. haematobium* in the experimental study. *Bulinus nasutus* from Unguja and Pemba were refractory to both Unguja and Pemba *S. haematobium* isolates (Table 2). This difference

Table 2 Summary of two experimental exposures to assess the compatibility of Schistosoma haematobium isolates to *Bulinus globosus* and *Bulinus nasutus* from Unguja and Pemba.

***Bulinus* sp. exposed**	**Snails collected from: Island (Shehia (s))**	***Schistosoma haematobium* isolate Island (Shehia)**	**Number of snails survived to shedding and number infected**	**References**
B. nasutus	**Unguja** (Kidimni Darajani – Koani Central Unguja and Kibonde Maji – Muyuni south Unguja)	**Unguja** (unknown)	17 survived **0 infected**	Stothard and Rollinson (1997a)
	Unguja (Kibonde Maji – Muyuni)	**Unguja** (Kinyasini)	43 survived **0 infected**	Stothard et al. (2000)
	Pemba (Vitongoji)	**Unguja** (unknown)	17 survived **0 infected**	Stothard and Rollinson (1997a)
	Pemba (Mtangeni)	**Pemba** (unknown)	9 survived **0 infected**	Stothard and Rollinson (1997a)
B. globosus	**Unguja** (Mdolo Pilau, Kinyasini, Kinsongoni Abdullah and swamp)	**Unguja** (unknown)	80 survived **37 infected**	Stothard and Rollinson (1997a)
	Unguja (Donge mbijii)	**Unguja** (Kinyasini)	24 survived **23 infected**	Stothard et al. (2000)
	Unguja (Mdolo Pilau, Kinsongoni swamp)	**Pemba** (unknown)	39 survived **5 infected**	Stothard and Rollinson (1997a)
	Pemba (Kimbuni, Wingwi, Machengwe, Kwapeweza, Ngwachani)	**Unguja** (unknown)	60 survived **3 infected**	Stothard and Rollinson (1997a)
	Pemba (Kimbuni, Wingwi, Kwapeweza, Chanjamjawiri, Kipapo, Ngwachani)	**Pemba** (unknown)	93 survived **0 infected**	Stothard and Rollinson (1997a)

in *B. globosus* and *B. nasutus* susceptibility on the Zanzibar islands was shown not to be due to discrimination of the snails by the parasite (Allan et al., 2009, Allan et al., 2013). Although not tested on neonates or young-juveniles of *B. nasutus*, which could show a higher susceptibility to schistosome infections and perhaps offer a subject for future research, it was concluded in the 2000s that *B. nasutus* was refractory to infection, not simply as a function of exposure, but due to genetically modulated factors of the *B. nasutus* innate immune system (Stothard et al., 2002c, Allan et al., 2009). Although this has since been contradicted with the finding that wild caught *B. nasutus* can be infected with *S. haematobium* (see below), the results of these experimental infections meant that large-scale control and research into schistosome transmission, which was again being considered at the turn of the 21st Century, could be concentrated in regions of active transmission, i.e. where *B. globosus* occurs (Stothard et al., 2002d, Knopp et al., 2013a).

Under the premise that *B. globosus* was the sole intermediate host for *S. haematobium* on the Zanzibar islands as detailed above, interest was given to comparing the modern day distribution of *B. nasutus* in regions, particularly southern Unguja, that had historically a high prevalence (Goatly and Jordan, 1965). Baseline *S. haematobium* epidemiological data collected from schoolchildren from Mtende school, located in a *B. nasutus* endemic region in southern Unguja, revealed a single heavy intensity infection in a student, who reported travel to northern Unguja but reported not to have bathed in any freshwater where *B. globosus* may have been present (Stothard et al., 2002d). The role of *B. nasutus* as an intermediate host for *S. haematobium* was questioned further when positive results from a diagnostic *S. haematobium* molecular marker were observed in snails suspected to be *B. nasutus* from Kangagani, Pemba (Ame, 2018) In follow-up surveys, a single naturally infected (shedding *S. haematobium* cercariae) *B. nasutus* (species determined using molecular markers) was recovered from Pemba in 2019, it was found transmitting a 'Mainland Africa' group one (Webster et al., 2012) strain of *S. haematobium* rather than the 'Indian Ocean Islands' group two (Webster et al., 2012) strain commonly observed on the Zanzibar islands (Pennance et al., 2022a). It seems plausible that variation in the genetically determined host-parasite specificity, which occurs on Unguja and Pemba between *S. haematobium* and *B. nasutus*, may partially account for previous observations of schistosome infections being acquired in *B. nasutus* habitats (Stothard et al., 2002d), although this does seem to be an infrequent occurrence (Pennance et al., 2022a). For example, despite the collection of *Bulinus* from *B. nasutus* habitats during ZEST, no infected

B. nasutus were collected from these habitats, and the 44 naturally infected among nearly 10,000 snails collected were all identified as *B. globosus* (Knopp et al., 2019c, Allan et al., 2020, Pennance et al., 2022a). However, a recent study by He et al. (2019) reported schistosome infected *Bulinus* from regions that are known to be *B. nasutus* habitats, and areas that have a historically high human infection prevalence (Vitongoji and Uwandani) (Knopp et al., 2019b, Pennance et al., 2022a). Although neither snail nor schistosome were identified to the species level using the necessary molecular markers, they were infected with schistosomes, which could also represent *S. haematobium* transmission through *B. nasutus*. It is apparent that, for the avoidance of doubt, further sampling of *B. nasutus* from endemic areas needs to be undertaken, and the species of both snail and schistosome inferred through molecular markers. It should also be considered that low levels of infection may be sustained through imported infections, and not local transmission, as is the case on Mafia Island (Tanzania) (Stothard et al., 2013), demonstrating the importance of collecting data on mobility of individuals during epidemiological surveys.

Within the ZEST project, the most recent and largest malacological collection effort to date on the islands, it was found that <1% of *B. africanus* species group snails were actively shedding *S. haematobium* cercariae (Knopp et al., 2019b, Allan et al., 2020). However, this work was bound by study design to only collect snails in specific shehias (snail control intervention arm) and not in other regions with more freshwater habitats. During a study investigating persisting transmission hotspots on Unguja, a higher infection prevalence of *Bulinus* (2.3%) was recorded (Pennance et al., 2016).

The two cryptic species of the *B. forskalii* species group, *B. barthi* and *B. forskalii* sp. present on Unguja and Pemba, are closely related to *B. cernicus*, the sole intermediate host of *S. haematobium* on Mauritius (Rollinson and Wright, 1984), and *B. bavayi*, an endemic species to Madagascar that has been shown to be compatible with *S. haematobium* locally (Stothard et al., 2001). It is possible that *B. barthi* and *B. forskalii* sp. may have some capacity to act as an intermediate host for *S. haematobium* on Unguja and Pemba (Stothard et al., 2002b). However, experimental infection of undelimited species of the *B. forskalii* species group from Zanzibar with *S. haematobium* were later determined refractory following observed exposure (Stothard et al., 2013). The intermediate host status of these *B. forskalii* species group snails should be revisited with snails identified to the species level to provide further reassurance in directing snail control and other snail focussed efforts.

6.4 Incrimination of *Bulinus* as intermediate host(s) of *Schistosoma bovis*

In 2016, schistosomes shed from *B. globosus* collected in Kinyasini, Pemba, were identified as being the cattle parasite *S. bovis* (Pennance et al., 2018). *Schistosoma bovis* is known to have a broad intermediate host range utilising several *Bulinus* species, including *B. forskalii* in mainland Africa (Mutani et al., 1983). The distribution of *B. forskalii* species on Pemba (Fig. 5) overlap with the regions in which *S. bovis* has been identified (Pennance et al., 2018, Pennance et al., 2021). Studies are therefore necessary to confirm the intermediate host snail vectoral capacity of *S. bovis*, including the collection and shedding of *B. forskalii* and *B. nasutus* in malacological surveys to determine the potential spread of this emerging species in Zanzibar.

6.5 Monitoring of *Schistosoma* infections in *Bulinus* spp

Traditional monitoring of the *S. haematobium* infections of *Bulinus* spp. in Zanzibar has been complicated by the identification of *S. bovis* on Pemba, where *B. globosus* acts as the intermediate host species for both *S. haematobium* and *S. bovis* transmission (Pennance et al., 2018, Pennance et al., 2021). As morphological differentiation of *S. haematobium* species group cercariae to the species level is not possible (Frandsen and Christensen, 1984), there is a real need for molecular xenomonitoring and more careful identification of schistosome cercariae (Pennance et al., 2018). Rapid diagnostic assays that can accomplish accurate differentiation of *S. haematobium* and *S. bovis* from collected and preserved cercariae (Webster et al., 2010) or from snail tissue as part of a molecular snail xenomonitoring assay (Pennance et al., 2020b), are both robust and reliable methods that should be implemented in future surveillance. When the aforementioned snail xenomonitoring assay (Pennance et al., 2020b) was used between 2017 and 2018 on >5,000 snails collected from across Pemba, 2.7% and 0.2% of snails were identified to have an *S. haematobium* and *S. bovis* pre-patent infection, respectively, compared to 0.1% and <0.1% when only patent infections were considered (Pennance, 2020). The xenomonitoring assay was ~24 times more sensitive than standard parasitological methods of shedding snails, and demonstrates the feasibility, with a modest investment in reagents, for using xenomonitoring assays in elimination surveillance on Unguja and Pemba.

6.6 Habitat preferences of *Bulinus globosus* and *Bulinus nasutus*

Bulinus globosus and *B. nasutus* prefer either swampy pools or gently flowing streams, but may also be found in rice paddies, and both also inhabit temporal freshwater bodies (Forsyth and Macdonald, 1966, Mccullough & Krafft, 1976, Stothard et al., 2002c, Allan et al., 2013, Pennance et al., 2022a). Both species have a marked preference for the underside of common blue waterlily leaves (Fig. 7) or Papyrus stems but may attach themselves to any convenient foliage (Mansfield-Aders, 1928, Mccarthy, 1930).

Both Zanzibar islands provide an abundance of suitable habitats for *Bulinus* species. The physical geography (relative north position), topography (more hilly) and climate (increased rainfall) favour the presence of habitats for *Bulinus*, and other freshwater snails, on Pemba to a greater extent than on Unguja (Mozley, 1939, United Nations, 1987). In the north of Unguja, however, there is a concentration of permanent streams, swamps and ponds that are filled by both prolonged and heavier rains than are observed in the south (Stothard et al., 2002c), where in addition to less rain, more porous/dry and infertile coralline limestone is found (Hardy et al., 2015).

Seasonal weather patterns in Zanzibar profoundly influence the population structure of *Bulinus*, with extended periods of drought (June to October) risking desiccation [although note the capacity of *Bulinus* snails to

Fig. 7 A typical habitat for species of the *Bulinus africanus* species group in Pemba taken during a malacological survey in 2017, also demonstrating active use of a freshwater site for washing clothes (A). Snails can be easily collected from the underside of the waterlily leaves covering the ponds surface (B) (personal collection, Bonnie L. Webster).

withstand considerable drought periods through aestivation (Cawston, 1927a, Allan et al., 2013, Kalinda and Chimbari, 2022)] and heavy bursts of rainfall flushing snails from preferred habitats (Mccullough & Krafft, 1976). Collecting *Bulinus* during the rainy season can be difficult due to their displacement in the water column (that may also cause snail death) and the increased size of water bodies, which may severely confound accurate measures of true snail abundance.

Like the geological landscape of coastal East Africa to which Unguja and Pemba have underlying shared features that predate their later isolation from mainland due to the Indian Ocean channel (Kent et al., 1971, Prendergast et al., 2016), the geology is dominated by limestone forming karst landscapes (Hardy et al., 2015). Corrosion of calcium carbonate by water creates sinkholes, cave systems, divergence of rivers and streams underground that may resurface downstream to feed surface pools and streams and dolines (Ford and Williams, 2007, Hardy et al., 2015). Dolines, bowl shaped depressions that provide foci for local surface drainage, provide suitable habitats for *Bulinus* snails, as is evident along ponds on the east coast of Pemba (Pennance et al., 2022a). Because of the karst landscapes on Unguja and Pemba, surface topographic wetness, which is often used to predict schistosomiasis transmission sites, is an unreliable predictor to use for mapping surface freshwater.

Consideration of the faunal associations putatively linked with known geological zones were first given by Mccullough & Krafft (1976) before being explored further by Stothard et al. (2000). An interesting feature are the limestone (M1) outcrops in the south and central regions of Unguja (Figs. 5 and 6), which elevate the water table because of the makeup of surrounding denser rock, thereby increasing the chance of surface water (Hardy et al., 2015). Only *B. nasutus* are present in these limestone outcrops in southern Unguja, and one limestone outcrop in central Unguja as well as on the 'bedrock' (M3-Sandy, Clay Marl), whereas on Pemba the same species is restricted to the sliver of Q2 coralline limestone in the eastern part of the island (Fig. 6). In the case of *B. globosus*, this species dominates the whole range of the M1 limestone on Pemba, but on Unguja a less clear pattern is seen due to the mosaic of geological characteristics in the north of the island, with this species presence recorded in three different geological zones (Fig. 6). *Bulinus forskalii* species group snails are widespread across multiple geological zones on both islands, although do not seem present in the south of Unguja (Fig. 5), perhaps due to these habitats being unfavourable for their survival, or due to a lack of

opportunity to colonise these regions through passive dispersal mechanisms, such as ectozoochory and endozoochory (i.e. the dispersal of snails through external attachment to another animals body or passing through another animal via internal consumption) (Simonová et al., 2016).

The mosaic of underlying geology on Unguja and Pemba has an impact on the physiochemical parameters of the freshwater. The conditions in relation to the survival of *Bulinus* on Unguja and Pemba were considered on a small scale first (Goatly and Jordan, 1965), but multiple studies since have investigated various physiochemical conditions, such as conductivity, pH, temperature, total dissolved solids and dissolved oxygen (Allan et al., 2013, Knopp et al., 2013a, Pennance et al., 2016), although none [except perhaps temperature, see Knopp et al. (2013a)] were significantly associated with predicting *Bulinus* spp. presence. Regarding specific preferences of *B. globosus* and *B. nasutus*, it was found that water conductivity was significantly lower in *B. nasutus* (albeit with one outlier) than *B. globosus* habitats (Stothard et al., 2000). However, preliminary results also showed that *B. nasutus* could survive in waters collected from *B. globosus* habitats (Stothard et al., 2000), and therefore it is unclear as to whether these parameters, or others, truly dictate *Bulinus* presence.

6.7 Interspecies and intraspecies genetic diversity of *Bulinus globosus* and *Bulinus nasutus*

Considerable intraspecies genetic heterogeneities within and between samples of *B. globosus* and *B. nasutus* from Zanzibar were observed first from RAPD analysis, where no two RAPD products from specimens were the same (Stothard et al., 1997), and nucleotide diversity of the partial *cox*1 showed multiple interspecies and intraspecies single nucleotide polymorphisms (Stothard and Rollinson, 1997a). Limited genetic variation within populations of *B. globosus* from multiple populations across Unguja and Pemba was observed from microsatellite loci analysis, however substantial genetic variation was observed between populations suggesting that gene flow was restricted, likely as a result of isolation by distance (Emery et al., 2003, Wilkinson et al., 2007). It was also observed that this genetic variation, and inbreeding, fluctuated temporally, demonstrating population expansion and contraction through bottlenecks caused by habitat desiccation during dry seasons (Wilkinson et al., 2007).

Snail populations from Unguja have been noted both from microsatellite, *cox*1 and complete mitochondrial genome/nuclear rDNA phylogenetic analysis to be more genetically similar to Kenyan and mainland

Tanzanian populations than those from Pemba (Wilkinson et al., 2007, Kane et al., 2008, Pennance, 2020, Pennance et al., 2022a). This likely reflects the earlier separation of Pemba from East Africa (~6 Ma) compared to Unguja (maybe as little as 10,000–18,000 years), where *Bulinus* population admixing would have continued for longer (Kent et al., 1971, Prendergast et al., 2016, Pennance, 2020). Interestingly, this is in contrast to the terrestrial molluscs of Pemba, where the difference in isolation times from Unguja are not reflected in the snail fauna (Rowson et al., 2010). Compared to *B. globosus*, it seems that *B. nasutus* has extremely low diversity across the Zanzibar islands as well as coastal Tanzania, despite potentially several millennia geographically separating these populations (Pennance, 2020). The taxonomic boundaries between species of the *Bulinus* genus in Zanzibar relative to those on other Indian Ocean islands and mainland Africa should be investigated by future population genomic studies.

6.8 Outlook

Many questions remain regarding the intermediate host snails of schistosomes on Unguja and Pemba. Snail surveys and genetic analyses of the snails and any *Schistosoma* species they may be infected with, provide a valuable resource to ongoing elimination efforts in Zanzibar. Perhaps the most pertinent area for ongoing research surrounds the compatibility of *B. nasutus* and *B. forskalii* species group to *S. haematobium* and *S. bovis*, since their status as potential intermediate hosts could have a significant impact on the surveillance of urogenital schistosomiasis and the potential spread of bovine schistosomiasis transmission. It is recommended that molecular snail xenomonitoring assays should involve all *Bulinus* species on the islands to assess the contribution of *B. nasutus* and *B. forskalii* species group to the current schistosome transmission dynamics. In addition, appropriate molecular markers to delimit snail and schistosome species, including hybrids, are required in future surveillance approaches. Further investigation is also needed to understand the ecological and geological factors that contribute to the distribution of the endemic *Bulinus* in Zanzibar. However, it is likely that developments in freshwater infrastructure to meet growing agricultural demands, such as the recently constructed dam in Kinyasini, Unguja and Pujini, Pemba (Fig. 8), will have the most dramatic impacts on *Bulinus* distribution on the islands. New waters establish suitable and larger habitats for freshwater snails thus increasing snail abundance and potentially favouring the introduction of invasive species, as is the case with

Fig. 8 Photographs from November 2021 of dam project in Kinyasini (Unguja, −5.9834438, 39.3060592) (A–F) and from May 2023 in Pujini (Pemba, −5.3172761, 39.7775620) (G–H), constructed for the Zanzibar Irrigation Infrastructure Project aimed at improving agricultural irrigation to boost local crop yield. Photographs show the

Pomacea canaliculata in Kenya (Buddie et al., 2021), the implications of which could affect *S. haematobium* transmission. The repercussions of increased schistosome transmission due to expanded intermediate host ranges jeopardise current targets for elimination of the parasite. Finally, the amount of work that has been carried out on snail-schistosome biology on Zanzibar offers an excellent foundation to build upon using next-generation technologies, geographic information systems and climate predictions. One has the capacity and tools to explore compelling questions such as the immunologic responses of snails to *Schistosoma* and the evolutionary history of these taxa in sub-Saharan Africa.

7. Molecular epidemiology

7.1 Genetic epidemiology of *Schistosoma haematobium* in Zanzibar

In the context of schistosomiasis control, the genetic diversity within and between *Schistosoma* populations provides an additional proxy to assess how control interventions may induce selection pressures, especially since the genetic diversity of the *Schistosoma* population may influence the outcome of control measures if variation is directly correlated to drug susceptibility/tolerance or with reduced/increased human host morbidity (Rollinson et al., 2009b).

Due to the geographical isolation of Unguja and Pemba from mainland Africa and their relatively small size, the Zanzibar archipelago provides a particularly suitable environment for assessing the impact of disease control strategies. The geographical features together with the long history of intensive schistosomiasis control interventions in Zanzibar, provide a unique opportunity to observe how *S. haematobium* populations respond genetically as they are pushed towards elimination.

A major evolution in molecular epidemiological methodologies for investigating *Schistosoma* populations, utilising DNA capture on Whatman

Kinyasini main dam wall (A), part of the Kinyasini reservoir viewed form the top of the dam wall (B), the irrigation canals leading from the Kinyasini reservoir to rice fields (C), the irrigated rice fields downstream (D), the suitable habitats for *Bulinus* spp. in the Kinyasini reservoir (E), collections of *Bulinus globosus* from the Kinyasini reservoir (F), Pujini dam reservoir showing irrigation channel (G) and the Pujini dam wall (H). (A, C, G, H: personal collection, Lydia Trippler; B, D, E, F: personal collection, Stefanie Knopp).

FTA cards (Gower et al., 2007) coupled with an alkaline elution protocol (Webster et al., 2013a) was demonstrated using larval forms of *S. haematobium* populations from Zanzibar. Prior to this technique, schistosome DNA could only be consistently captured from adult worms, therefore larval parasites had to be passaged through laboratory rodents and snails before assessing their genetics, introducing selection biases and population bottlenecks. By being able to directly sample natural *Schistosoma* populations via larval miracidia and cercariae from their human or snail hosts, respectively, it was now possible to analyse truly representative population genetics and genomics (Webster et al., 2012, Webster et al., 2013a). Without the development of this method, several important findings regarding *Schistosoma* populations such as detecting interspecies introgression/hybridisation and selection may not have been uncovered (Webster et al., 2013b, Léger et al., 2016, Platt et al., 2019, Pennance et al., 2020a, Rey et al., 2021).

Zanzibari *S. haematobium* populations have been analysed at several time points along the evolution of molecular tools and methodologies, starting from the analysis of small DNA regions including mitochondrial genes and nuclear rRNA regions (Webster et al., 2013a, Pennance et al., 2018) and genome wide microsatellite markers (Webster et al., 2015, Pennance et al., 2022a), to whole genome analyses (Platt et al., 2019). It is important to highlight that the studies investigating the genetics of *Schistosoma* populations in Zanzibar would not have been possible without the collection of both *Schistosoma* and intermediate host snails within multidisciplinary research programmes in Zanzibar. Sample collections, combined with the appropriate downstream collection maintenance and data documentation (including the appropriate collection and exporting permits) in repositories have facilitated molecular investigations: the value of these archived collections, in terms of capturing genetic material from specific times and places for subsequent reference, should be emphasised (Emery et al., 2012).

7.2 Genetic diversity and population structure of *Schistosoma haematobium* on Unguja and Pemba

The first genetic assessment of *S. haematobium* populations from four shehias in Unguja (Kiboje, Kilombero, Kinyasini and Mwera) and one shehia in Pemba (Wingwi) analysed two mitochondrial DNA regions, the *cox*1 and the NADH dehydrogenase subunit 1 (*nad*1) (Webster et al., 2013a). Additionally, the first and second ITS (ITS1 and ITS2, respectively) were analysed to determine any variation at the species level (Webster et al., 2012).

A significant finding from the study was that Zanzibari *S. haematobium* split into two very distinct groups. The net divergence between these groups was considerably high given the geographical scale of Unguja and Pemba, with similar levels usually seen between genetically diverse lineages of *S. mansoni* spread thousands of miles across Africa (Webster et al., 2013a). The observed groups were constant across the samples from the three time points (2001/2008/2009), indicating that separation was influenced by some level of evolutionary divergence. Interestingly, by comparisons with a small selection of other geographical strains of *S. haematobium*, Zanzibari *S. haematobium* together with other endemic Indian Ocean islands (Madagascar and Mauritius) formed a unique group (named group two) that contained no *S. haematobium* from the African mainland (Webster et al., 2012). The other genetic group (named group one) contained a cluster of *S. haematobium* from across mainland Africa together with some *S. haematobium* isolates from Zanzibar. These two groups of *S. haematobium* have also been identified as being transmitted by *Bulinus* snails on Unguja and Pemba, showing that *S. haematobium* of both groups can be acquired in Zanzibar and are not imported (Pennance et al., 2022a). This splitting of the two groups and the higher diversity of *S. haematobium* group two was further confirmed at the genomic level by Platt et al. (2019).

Contrary to the theory that island populations are less diverse because of their relative isolation and interbreeding, the group two *S. haematobium* cluster, and specifically Zanzibar isolates, show high levels of genetic diversity and a complex genetic network compared with those from the African mainland, which appeared monomorphic (Webster et al., 2013a). This higher genetic diversity was also confirmed using nuclear molecular markers in two pilot microsatellite studies (Glenn et al., 2013, Webster et al., 2015). It was hypothesised that the high level of genetic diversity of the *S. haematobium* populations was a result of importation of *S. haematobium* into Zanzibar via the Indian Ocean trade routes mainly from the Arabian Peninsula, which started possibly as early as the late 5th to 8th century Common Era (Crowther et al., 2015), with further spread of *S. haematobium* over to the African mainland. This came several thousand years following the first known presence of urogenital schistosomes in the region, with eggs being found in 6,200 year old human skeletal remains from Tell Zeidan, Syria (Anastasiou et al., 2014). This theory also fits the genetic diversity observed and the low-level mixing of *S. haematobium* populations from Zanzibar and the mainland East African coastal regions, namely Tanzania and Kenya. A study that analysed *S. haematobium*

populations from Yemen supports the hypothesis that *S. haematobium* was imported mainly from the Arabian Peninsula; with samples from Yemen showing high levels of diversity within the population, as with Zanzibar, but with observable relatedness in *cox*1 haplotype networks between these two *S. haematobium* populations (Sady et al., 2015). Furthermore, the clustering of *S. haematobium* collected from infected individuals from Mafia Island (Tanzania) with the group two *S. haematobium* from Zanzibar helped elucidate that those human *S. haematobium* infections on Mafia Island were imported cases from Zanzibar (Stothard et al., 2013).

7.3 Do intense control interventions change the composition of *Schistosoma haematobium* populations?

Through routine schistosome infection surveillance, it is quite possible to observe, or estimate, changes in terms of the abundance of parasites in human or snail populations when targeted with control interventions. However, to determine if selection pressures are imposing themselves on parasites differentially, perhaps due to advantageous genotypes/phenotypes, requires other measures. The study by Webster et al. (2013a) used the mitochondrial data, generated from *S. haematobium* populations collected on Unguja and Pemba in 2001, 2008 and 2009, to investigate any population level effects of the MDA programmes. Between 2001 and 2008/2009, MDA programmes had been intensified on both Unguja and Pemba (see section Treatment interventions) increasing any praziquantel selection pressures on the *S. haematobium* populations. Although from a relatively small sample size, the similarity of *S. haematobium* mitochondrial haplotypes from 2001 to 2008/2009 suggested that, despite large-scale MDA, there had been no direct impact on the parasite populations or any indication of selection of specific *S. haematobium* genotypes in Zanzibar.

The impact of intensified MDA (biannual treatment) and also other interventions, such as snail control or behaviour change interventions during ZEST, on *S. haematobium* populations was specifically investigated as part of the parasite population genetic component of SCORE (Webster et al., 2020). Analyses were conducted using optimised panels of genome wide nuclear microsatellite markers, which were developed as a tool to enable affordable and high throughput genotyping of *S. haematobium* miracidia (Glenn et al., 2013, Webster et al., 2015). Just over 3000 miracidia collected from infected individuals on Unguja and Pemba before the onset of ZEST interventions in 2012 and again in 2016 were analysed (Pennance et al., 2022b). These collections bookended three years of

biannual MDA (2012–2015) and additional elimination targeted interventions (see Introduction). The microsatellite data were analysed to (1) assess the population genetic composition of the *S. haematobium* populations over space and time and (2) to investigate any differential impact that the ZEST interventions may have had on the *S. haematobium* populations. In areas where MDA and snail control had been implemented, a greater reduction in both the *S. haematobium* populations genetic diversity and inbreeding coefficient (increased outcrossing) was observed despite there being no difference in infection epidemiology (prevalence and intensity) in relation to the different interventions. Additionally, using a statistical analysis developed to estimate adult worm fecundity (egg output) (Neves et al., 2021), it was proposed that on Pemba, *S. haematobium* female worms had greater egg output than those on Unguja. It was hypothesised that this may indicate a strain dependent phenotype of the *S. haematobium* populations possibly driving persistence of transmission on Pemba, although further sampling would be required to elucidate this. Moreover, no biological difference was observed in *S. haematobium* populations circulating in persistent infection hotspots compared with those in areas where infection prevalence was low and decreasing over time.

7.4 Emergence of *Schistosoma bovis* in Zanzibar

One of Zanzibar's unique epidemiological traits was that it was originally endemic for only *S. haematobium,* whereas for many other schistosomiasis endemic countries, multiple *Schistosoma* species are transmitted. However, in 2016, via a widespread and detailed xenomonitoring programme conducted across Pemba (Pennance, 2020), five *B. globosus* were found to shed *S. bovis* cercariae, at a single locality in the north of the island (Pennance et al., 2018). In 2019, *S. bovis* infections were confirmed in cattle grazing at the same site the infected snails were found by the molecular identification of miracidia retrieved from stool samples (Pennance et al., 2021). Mitochondrial *cox1* analysis of the *S. bovis* cercariae and miracidia suggested that the origin of the *S. bovis* was the East African mainland, probably through cattle importation, which increased considerably during the late 20th Century (Pennance et al., 2021). Further analysis of the snail collections made during the xenomonitoring programme, using a pre-patent xenomonitoring assay (Pennance et al., 2020b), indicated a wider distribution of *S. bovis* on Pemba (Pennance, 2020). These studies confirmed that *S. bovis* transmission is now established on Pemba, facilitated by susceptible *Bulinus* host snails. The active transmission of *S. bovis* poses a risk to livestock

health on Pemba, which also has an economic impact, with further surveillance warranted (Pennance et al., 2021). Furthermore, hybridisation between *S. haematobium* and *S. bovis,* as observed on the African mainland could change the disease epidemiology in Zanzibar, increasing transmission through other *Bulinus* species and potentially creating the risk of zoonoses. However, recent genomic investigations of Zanzibar isolates have not shown a genetic interspecies introgression with Zanzibari *S. haematobium*, which is considered to be the purest form of the species (Platt et al., 2019).

The establishment *of S. bovis* complicates transmission monitoring of *S. haematobium* via snail xenomonitoring approaches, since not only sensitive but also species-specific markers are needed for accurate determination of human and/or animal schistosomiasis transmission. This will be particularly important for surveillance for *S. haematobium* elimination (World Health Organization, 2022).

7.5 Outlook

The use of bespoke larval (cercariae and miracidia) schistosome collection methods, together with the associated genetic analyses, have enabled insights into the epidemiology and genetic diversity of the natural *Schistosoma* populations over space and time. Subtle genetic differences undoubtedly exist in Zanzibari *S. haematobium* populations, whilst the potential population level associated phenotypic differences observed, certainly warrant further investigations.

It remains unknown if these unique genetic traits are related to differences in transmission and disease epidemiology, however, it is certainly clear that Zanzibari *S. haematobium* populations are not comparable to those from the African mainland. Increased sample sets and population genomic level analyses (Shortt et al., 2021) would provide further insights into these genetic traits and if they impact control interventions.

The detection of *S. bovis* transmission on Pemba highlights the need for detailed molecular identification of *Schistosoma* samples, particularly from snails. This finding, although small, demonstrates the need for more in-depth surveillance methodologies going forward, whilst highlighting the fact that schistosomiasis is a complexed dynamic disease. The concern now is that *S. haematobium-bovis* hybrids will soon be transmitted in Zanzibar, although how this may affect ongoing control efforts remains unknown. As on the African mainland, genomic surveillance of *Schistosoma* populations is needed to fully understand the dynamics of these hybridisation events, together with any impact on disease epidemiology and control.

(Lund et al., 2022). Finally, with the identification of a mechanism of action of praziquantel and the genetic mutations that can infer drug resistance in schistosomes (Park et al., 2019, Le Clec'h et al., 2021), population level genomic surveillance could support the monitoring of any drug selection pressures on the Zanzibari *S. haematobium* populations, as they are pushed towards elimination.

8. Diagnostics

Over the last ~56 years, from 1966 to the present day, the literature shows an evolution in the diagnostic methodologies used and/or tested in Zanzibar (Table 1). Reports range from small-scale basic descriptive diagnosis to large-scale diagnostic evaluation and implementation; from application of simple point of care tests to assess morbidity and monitor the impact of control interventions, to the development of innovative advanced assays needed to certify and sustain elimination and implement post-elimination surveillance. It is no surprise that the early publications and reports focus on basic diagnostics such as haematuria detection and microscopy, whereas the more recent publications and reports present more advanced diagnostics such as antigen and molecular tests.

8.1 First diagnostic research in Zanzibar

The first diagnostic research study conducted in Zanzibar was published in 1966 and was performed to assess bladder morbidity associated with urogenital schistosomiasis (Forsyth and Macdonald, 1966). The study took place in Donge school on Unguja where stool and urine samples were collected from 517 children. Ten millilitres of each urine sample, collected between 12 and 2 pm, were centrifuged and the deposit was examined for the presence of *S. haematobium* eggs. If the sample was egg-positive, a second 10 mL sample was taken from the participant, and the number of eggs found was counted to provide an intensity score. Urine analysis was conducted on three consecutive days and only children who had no eggs in all three urine samples were recorded as negative for infection. A clinical examination was also carried out, including height, weight, blood tests, electrocardiograms to detect hypertension and urography (X-ray image of the renal pelvis and urinary tract). A total of 45.3% of the children were found to be egg-positive and the urography provided interesting insights into the related urogenital pathology (see section Morbidity). Stool samples

were mainly analysed to determine soil-transmitted helminth infections, however eight individuals were also found to be passing *S. haematobium* eggs in their stool. It was hypothesised that this observation was attributable to high intensity *S. haematobium* infections with spill-over of eggs into the intestinal tract (Cunin et al., 2003), although this can also be a result of contamination of faecal samples by the patient's urine during sample taking. Additionally, two children were found to have intestinal schistosomiasis (*S. mansoni*) despite reporting to have never visited the African mainland. In the publication, this did not lead to any speculation that there was any transmission of *S. mansoni* in Zanzibar. A *S. mansoni* infection was also identified in a later study, via stool examination in a study on Pemba in 1984 (Pampiglione et al., 1987), but the adult male had a history of travel to the African mainland and so this was recorded as an imported case.

Following on from earlier investigations by Forsyth and Macdonald (1966), a longitudinal study of 1,074 participants from four villages on Unguja was conducted over two years in an attempt to document urological abnormalities caused by *S. haematobium*, how they progress and how they affect the patient's health and well-being (Forsyth, 1969). *Schistosoma haematobium* infection was determined by examination of urine sediments, as described in the earlier study (Forsyth and Macdonald, 1966), with a 65.1% prevalence recorded. This diagnostic procedure allowed for a good assessment of the general health and urological abnormalities of infected individuals of varying ages with different intensities of infection.

8.2 Immuno-diagnosis-antibody detection

Forsyth (1971) also led the first investigation of two novel immunological diagnostic approaches in Zanzibar: the intradermal and the plasma card test. These tests were compared with egg detection in urine sediments to ascertain their reliability in a setting such as Zanzibar (Forsyth and Macdonald, 1966). The intradermal test (Pellegrino, 1958) involved the injection of *Schistosoma* antigen (prepared from cercariae, eggs, adult worms or miracidia) under the skin of participants' forearms. A positive diagnosis was observed by the development of an immune reaction seen as a hive (raised rash), referred to now as a wheal, of $>1\ cm^2$, with the size of the wheal related to the intensity of infection. However, wheals were not often uniform in their shape. The plasma card test (Sadun et al., 1963), involved obtaining serum from three drops of blood from the patient, which was then mixed with *Schistosoma* antigen on test cards originally developed for the diagnosis of syphilis and treponematoses (Portnoy et al., 1962).

Clumping of the plasma sample on the card provided a positive diagnosis of schistosomiasis. A total of 290 patients were tested using the intradermal skin test, 87 patients were tested using the plasma card test, and a subset of 57 patients were tested using both the intradermal test and the plasma test (Forsyth, 1971). The data from this study showed a high prevalence (83.4%) of infection by urinary egg microscopy, which was used as the benchmark/gold standard for testing sensitivity of the new tests. The intradermal skin test fell short of detecting all positive cases, with an overall sensitivity of 76.4%, with the largest proportion of false negative test results being from young children (0–5 years) where sensitivity dropped as low as 43.8%. The plasma card test, however, was reported to have a high sensitivity of 100% in young children (0–5 years). This dropped considerably in other age groups, resulting in an overall sensitivity of 76.3%. Whereas no association was found between the positive skin test result and the intensity of infection (as assessed by urine microscopy), a positive plasma test result was usually associated with a higher intensity of infection, demonstrating that sensitivity of the plasma test likely increased in higher intensity infections. Of the 57 patients that were tested with all three diagnostics, only 20 (35.1%) of the patients tested positive for both the skin and plasma test, although 48 (84.2%) were egg-positive by urine microscopy. It was concluded that the intradermal and plasma card tests were not sensitive immunodiagnostic tests for use in Zanzibar.

Further immunodiagnostic research was carried out in 1990, as part of the 'test and treat' strategy within the Schistosomiasis Control Programme (Xue et al., 1993). This involved the use of haemocyanin, obtained from the keyhole limpet (*Megathura* crenulata), which contains the same carbohydrate as that found on the surface of *S. mansoni* schistosomula (Grzych et al., 1987). A total of 187 serum samples were collected from individuals on Pemba and used to compare the keyhole limpet haemocyanin enzyme-linked immunosorbent assay (ELISA) to the standard soluble egg antigen (SEA) ELISA (Xue et al., 1993). Compared with haematuria and/or egg-positivity, it was found that the sensitivity of the tests were high, but specificities were low. Although useful, these immunological tests added little value in highly endemic settings like Zanzibar (at that time) and the new biomarkers did not perform to a high standard.

Other studies have tested alternative immunodiagnostic methods such as indirect haemagglutination (IHA) and western blot assays, which showed a high prevalence of exposure to *S. haematobium* within Zanzibar (Bevilacqua et al., 2012). Western blot assays proved to be a useful tool for

assessing seroprevalence for urogenital schistosomiasis but the IHA assay showed high cross reactivity for other helminths.

In 2008, the standard SEA ELISA was tested on serum samples from 150 children on Unguja, showing that this immunological test was sensitive (89.0%) and specific (70.0%) (Stothard et al., 2009c). Further work confirmed the high sensitivity of SEA ELISA in Zanzibar with seroprevalences several-fold higher than indicated by egg detection diagnostics (Knopp et al., 2010). However, the main reason for this result is likely due to antibodies from past infections that remain for a prolonged time after successful treatment.

The high sensitivity of these immuno-diagnostics for schistosomiasis has little value in high endemicity settings, as they cannot discriminate between current and past infections (Doenhoff et al., 2004, Weerakoon et al., 2015). However, it is acknowledged that immuno-diagnostics will soon be needed in settings like Zanzibar, once interruption of transmission is close. These types of tests will be needed as a monitoring and post-elimination surveillance tool in settings where a very low prevalence has been achieved, transmission is reduced and where only low levels of the communities are exposed to *S. haematobium* (Utzinger et al., 2015). In light of this, Pearson et al. (2021) recently focused on the selection and screening of new antigens for improved immuno-diagnosis. The study took advantage of the unique repository of urine samples collected and preserved during the ZEST project and stored on Pemba (Knopp et al., 2012, Knopp et al., 2019c). The ZEST sample repository at PHL-IdC in Pemba contains around 2,500 urine samples (10 mL) from adults, 6,500 urine samples (10 mL) and 37,000 urine samples (1.5 mL) from schoolchildren. All samples are frozen at −20 °C. In the study of Pearson et al. (2021), a total of 992 biomarkers were identified through analysis of an annotated *S. haematobium* genome and through orthologue comparisons, and screened with antibodies from the stored urine samples. Two potential biomarker candidates were identified in two tetraspanin type proteins, Sh-TSP-2 and MS3_01370, which were later incorporated into pilot point of care immunochromatographic tests. Both candidates exceeded the sensitivity of the routine SEA ELISA, with a 100% specificity, and presenting 75.0% (Sh-TSP-2) and 89.0% (MS3_01370) sensitivities. These biomarker candidates, when incorporated into the portable rapid diagnostic test (RDT) format, showed great potential for supporting schistosomiasis control and elimination initiatives, particular focusing on post-elimination surveillance.

8.3 Haematuria-based diagnosis

Urine haematuria used as a proxy marker for *S. haematobium* infection and associated morbidity, has been utilised to assess the impact and success of control programmes in Zanzibar from 1976 until this day (Table 1) (Mccullough, 1978, Trippler et al., 2021b).

A travel report from 1983 describes the implementation of the third parasitological survey of the Zanzibar Control Programme in the shehia of Kinyasini, Pemba (Mott et al., 1983). The report clearly details the assessment of the diagnostics used and provides recommendations for improved data recording. The routine diagnostics used were the quantitative urine filtration method (eggs/10 mL of urine) and macro- and microhaematuria, with the latter being used to assess levels of morbidity. It was recommended that a more detailed assessment of haematuria should be made, including history of haematuria, presence of bright red or cloudy urine and using a haematuria scoring system (negative, +, ++, +++), for increasing amounts of blood relating to the colour change on the urine reagent strip. This was the first suggestion for quantifying haematuria levels to support more targeted control efforts. Interestingly, this report details the need for the standardised use of urine reagent strips for haematuria assessment as clear discrepancies in performance were observed between different types of urine strips and manufacturers.

Following on from the travel report, a proposal for the evaluation and extension of the schistosomiasis control programme in Zanzibar submitted to the WHO in 1984 (Mgeni, 1984b), recommended that on Unguja, due to the resources available, quantitative urine filtration could be used throughout the programme. Surveys were to be conducted every six months with a final evaluation performed, at the end of the final year, using quantitative urine filtration to also assess intensity levels. In contrast, due to the lower diagnostic resources available on Pemba, it was suggested that haematuria should be used as the main assessment tool to monitor infection prevalence for the whole population over the remaining two years of the programme. The assessments were aimed at obtaining prevalence data for both islands to investigate whether the implemented interventions would be able to reduce infection prevalence and high intensity infections (>50 eggs/10 mL of urine) by 50% and 90%, respectively, within two years, and by 99% in five years. From these reports, it is clear that haematuria assessment was considered an informative tool for monitoring schistosomiasis infections as interventions start to have an impact.

8.4 Using haematuria as a proxy for treatment and sign of morbidity reduction

The first long-term schistosomiasis control programme in Zanzibar was initiated in 1986, with an aim of eliminating urogenital morbidity (Savioli et al., 1989b). Over the course of the programme, haematuria was further assessed as a proxy diagnosis for urogenital schistosomiasis. In 1989 to 1990, three papers were published that reported the first large-scale diagnostic projects in Zanzibar centred around the use of Hemastix reagent strips (Fig. 9) as a diagnostic tool to aid targeted treatment with an aim to reduce urogenital schistosomiasis-associated morbidity and transmission (Savioli et al., 1989b, Savioli and Mott, 1989, Savioli et al., 1990b).

A pilot study to assess the methodology was conducted in July 1988 in the village of Pujini on Pemba (Savioli et al., 1990b). At this time, no previous prevalence assessment or treatment had been done in this village. Daily urine samples from 879 individuals were examined on six consecutive days and each sample was examined using multiple methods: (1) visually for macrohaematuria and then (2) for microhaematuria using the Hemastix reagent strips and (3) by quantitative urine filtration. The Hemastix reagent strips results were measured in a scoring range from light (+) to heavy (+++) for each sample and interpreted by the strength of the reagent strips colour change from orange to green. The main findings from this study showed that egg excretion from infected individuals was highly variable from day to day, with some individuals excreting >50 eggs/10 mL of urine on one day and then being egg-negative on another. Macrohaematuria was shown to be 100% specific when compared with egg excretion and was closely associated with egg counts >50/10 mL. Macro- and microhaematuria were strongly correlated with egg excretion although there were some cases where individuals were egg-positive but had no trace of haematuria. This study provided strong evidence that at the primary health care level, single semi-quantitative measurers and obvious haematuria were more useful indicators of infection compared to a single egg count by microscopy. These methods were advocated as suitable surveillance tools that should be used in high prevalence settings to support treatment of individuals and to enable implementation of appropriate treatment strategies.

Due to the pilot study success, the haematuria-based methodology was implemented in a survey of >20,000 children from 52 schools across Pemba (Savioli et al., 1989b). An initial survey of 24,462 children took

Fig. 9 Diagnostic procedures being performed, barcoded urine samples are prepared through urine filtration on barcoded glass slides (A), followed by egg microscopy (B). Hemastix reagent strips are a useful tool for the detection of microhaematuria in infected individuals (C) and are more sensitive than circulating cathodic antigen (CCA) tests that can result in negative [single band on lateral flow test (D)] in many egg-positive samples that were detected through microhaematuria as shown by a green colour change on the Hemastix reagent strips of the same sample (D) (A: personal collection, Sophie Welsche; B, C: personal collection, Lydia Trippler; D: personal collection, Russell Stothard).

place in November 1986 with urine samples from each child being examined for macrohaematuria and microhaematuria. If macrohaematuria was observed, treatment of that individual took place without any further testing (test-and-treat) (see section Treatment interventions). If no macrohaematuria was observed, urinalysis Hemastix reagent strips were used to test for microhaematuria and treatment was provided if positive. The whole cohort of children were followed up six months later with further haematuria assessments. Compliance in this test-and-treat strategy was 99.0%, with an 84.9% reduction of haematuria observed. The results indicated that treatment had dramatically reduced infection and intensity levels, and also associated morbidity. The study truly demonstrated the feasibility of using indirect techniques (namely haematuria assessment) at a large scale to diagnose urogenital schistosomiasis, identifying children that require treatment, whilst also monitoring the impact of the interventions on clinical morbidity in a highly endemic area (Savioli et al., 1989b). The intervention objectives were to reduce macrohaematuria prevalence by 90% and microhaematuria prevalence by 65% within two years (Savioli and Mott, 1989). This objective was achieved, with macrohaematuria reduced by 94.5% and microhaematuria reduced by 69%, after only one year. Additionally, it was observed that the entire test-and-treat kit could be easily transported in a basket on a bicycle, motorbike or on foot, with a capability of the teams, which included five rural health assistants, to test-and-treat 26,000 individuals in one month. It was pointed out that it would take six months to screen this number by quantitative urine filtration. Following the findings, these test-and-treat approaches were rolled out across Pemba to reach all at-risk populations. However, it was noted that despite the successes of this intervention strategy, sensitivity and specificity of the methods needed continuous evaluation and as intensity and prevalence levels dropped, more sensitive and specific methods would need to be employed to detect low level infections. Indeed, the success of this methodology continued into 1991, where during a visit to Pemba by Muchiri (Division of Vector-Borne Diseases, Kenya), it was clear that haematuria had been kept to low levels, <25% (Muchiri, 1991). However, Muchiri did state in his report to the WHO that a dedicated team was needed to sustain morbidity at this low level and to further reduce the burden of urogenital schistosomiasis.

Following on from the test-and-treat campaign implemented within the Schistosomiasis Control Programme, further evaluation was done to investigate the sensitivity and specificity of indirect haematuria diagnostic

methods in relation to declining intensity and prevalence of infection (Savioli et al., 1990b). Data from the large-scale surveys were evaluated showing that microhaematuria, detected at the light positive (+) level presented the highest diagnostic sensitivity but the lowest specificity, whereas macrohaematuria had the highest specificity but the lowest sensitivity. Moderate microhaematuria (++) detection had high sensitivity and specificity levels, but these were still below the scores for quantitative urine filtration. The analyses suggested that the quantitative indirect methods tended to decrease in specificity during the course of the intervention programme, but that sensitivity remained relatively stable. Overall, the findings indicated that a history of haematuria could be used as preliminary screening methods to identify those at risk of morbidity within a population. Whereas, surveillance for light microhaematuria (+) is likely a more appropriate method for supporting targeted interventions at the individual level (Lwambo et al., 1997a).

Further data evaluation indicated that the prevalence of microhaematuria was linearly associated with the prevalence of infection, with macrohaematuria being a marker of infection intensity and morbidity risk in the community. These correlations appeared to stay consistent throughout repeated community-based treatment, suggesting that haematuria quantification was useful for assessing interventions and as a tool for long-term surveillance (Lwambo et al., 1997b).

The sensitivity of Hemastix reagent strips was also investigated to assess the applicability of expired Hemastix reagent strips batches for programme use (Savioli et al., 1993). When compared with an unexpired batch, reagent strips that were expired for one year but had been stored in their original sealed glass containers at room temperature (26–32 °C) on Pemba showed a sensitivity of only 59.7%. There were no false positives and a quarter (10/40) of samples that were haematuria-positive using unexpired Hemastix reagent strips, had less intense colour reactions than the expired Hemastix reagent strips, which resulted in a decreased microhaematuria grading according to the manufacturers manual (i.e. from +++ to ++ and from ++ to +). Only 39.5% (17/43) of the urine samples that were indicated as trace or light positive with the unexpired Hemastix reagent strips, were also haematuria-positive when the expired Hemastix reagent strips were used. As a consequence of these results the authors recommended that expired Hemastix reagent strips should not be considered for diagnosis of patients and for selective population treatment in schistosomiasis control programmes.

Almost 20 years later, in 2007, results of a longitudinal study were published that evaluated the use of microhaematuria assessment within ongoing control programmes, as a tool for tracking the dynamics of infection in the population and also for assessing the impact of anthelminthic control (French et al., 2007). At a cost of £0.20 per test, the use of reagent strips for the detection of microhaematuria enabling the monitoring of infection prevalence during ongoing control programmes was advocated as a rapid, cost-effective and informative surveillance tool.

Although used in several other small-scale studies for prevalence mapping and morbidity assessment (Stothard et al., 2002d), the use of haematuria as a diagnostic marker was not further evaluated in terms of its sensitivity and specificity until 2018 (Knopp et al., 2019a). In the meantime, Zanzibar was considered an elimination setting and the use of microhaematuria detection in relation to very low intensity infections was investigated. With a very large sample set (39,207 samples from children and 18,155 samples from adults), the study showed that there was a significant decrease in the diagnostic sensitivity of urine haematuria, as egg counts decrease. This was particularly evident for ultra-low infections (<5 eggs/10 mL of urine) where a mean sensitivity of only 52.0% was achieved. At the time of this study, after several intense rounds of MDA and other interventions within the ZEST project, the largest proportion of urinary egg counts were in the ultra-low category (34.7% and 46.7% of the children and adults, respectively). These ultra-low infections impose a major challenge for accurate diagnosis, with the use of haematuria detection not providing a sensitive measure. It was stated that next-generation diagnostics were needed in these ultra-low endemicity settings where interruption of transmission is the main objective, and where reliable detection of infections with <5 eggs/10 mL of urine is necessary to treat individuals that continue to contribute to transmission.

8.5 Urine analysis and urology as a diagnostic and morbidity marker

In the early 2000s, there were several studies conducted mainly on Unguja that focused on understanding the levels of urological morbidity associated with *S. haematobium* infection. Some of these morbidity markers, facilitated by various commercially available platforms and reagents, were also investigated in terms of their ability to provide a diagnosis for active urogenital schistosomiasis (Rollinson et al., 2005, Sousa-Figueiredo et al., 2009, Stothard et al., 2009b). In all three of these studies, morbidity

markers were assessed against the routine methods, macro- and microhaematuria, urine turbidity and quantitative urine filtration.

In 2004, a study explored the relationship between *S. haematobium* infections and excreted urine-albumin and urine-haemoglobin, using two existing rapid assays commonly used within healthcare systems for urine analysis (Rollinson et al., 2005). For this purpose, urine samples from 305 children were analysed, yielding a *S. haematobium* infection prevalence of 53.9% by quantitative urine filtration. Urines were also analysed using the HemoCue Urine Albumin assay for levels of microalbuminuria and the HemoCue Plasma/Low Hb assay for levels of haemoglobin. The data from the urine-haemoglobin assay clearly showed that quantification of excreted haemoglobin was possible and potentially useful. However, the HemoCue assay was much less sensitive than microhaematuria detection using Hemastix reagent strips. Interestingly, within this study, it was found that grading gross haematuria within urine samples, even using a standardised colour chart, was highly subjective with quality control related to how different individuals see/interpret colour. Turbidity was found to be a more sensitive and robust method for grading urines compared to colour. The HemoCue Urine Albumin assay, for determining levels of microalbuminuria as a proxy for *S. haematobium* infection, was considered useful as a rapid operational field diagnostic tool. Microalbuminuria levels were found to correlate with intensity of infection levels with a diagnostic sensitivity of 90.0%. However, it is unknown how this sensitivity may reduce with low intensity infections.

A study conducted in 2005 analysed multiple urine parameters, including levels of albumin measured using simple Microalbustix reagent strips, to identify morbidity, together with routine haematuria and urinary egg microscopy (Sousa-Figueiredo et al., 2009). In addition, a portable ultrasound machine was used to assess upper and lower urinary track morbidity, scored according to the WHO standardised protocol (Scientific Working Group On Schistosomiasis, 2006). Raised urine-albumin was associated, but not significantly, with prevalence of infection within the cohort, whereas microhaematuria was significantly associated with prevalence. Albuminuria was positively associated with *S. haematobium* infection showing a sensitivity and specificity of 63.3% and 83.1%, respectively.

A further study conducted in 2006 showed complimentary results (Stothard et al., 2009b). Urine samples from 66 children were collected over three consecutive days. Egg patent infections were at a prevalence of

65.2%, haematuria was recorded for 77.3% of the participants and 66.1% had urinary tract pathology. Abnormal albumin/creatinine ratios were detected in 88.4% of the children, but no clear correlation with *S. haematobium* infections was identified.

In summary, these studies showed that albuminuria assays, particularly the use of urine dipsticks formats, are good and feasible diagnostic tools for use in endemic settings and are less laborious compared to microscopic egg detection. Additionally, they could be useful rapid morbidity assessment tools for monitoring *S. haematobium* related urine pathology, as well as a method for selecting individuals that have more chronic urinary lesions without having to conduct an ultrasonography assessment. However, the tests do not seem to have any diagnostic advantage over haematuria detection and the assessment of urogenital tract pathology. Measures of albumin, other urine parameters and urological pathology by ultrasound were suggested to be better applied as pre-screening tools enabling selection of schoolchildren for further investigations, whilst excluding those not needing any follow-up.

8.6 Immuno-diagnosis; antigen detection

In 2003, a novel lateral flow cathodic circulating antigen (CCA) dipstick ('schistosomiasis one step test') became commercially available for research diagnostic purposes. This test followed on from the original CCA developments by van Etten et al. (1994) and Van Dam et al. (2004), who aimed at producing a RDT that was storable, portable, easy to use and compatible with urine. The performance of the novel CCA RDT was evaluated on Unguja in June 2003 (Stothard et al., 2006a) on urine samples collected from children from two primary schools (Ghana and Mwera). The mean prevalence of *S. haematobium* across the two schools, based on quantitative urine egg microscopy, was 68.5%. Urines from 25 participants were selected for the CCA RDT evaluation. These urines ranged from being egg-negative, and from low to high egg counts. Urines were also graded for macro- and microhaematuria. Given that only *S. haematobium* is present on the Zanzibar islands, the study produced conclusive evidence that the CCA-based diagnostics were not suitable for urogenital schistosomiasis assessment and that CCA was not detectable for *S. haematobium* in Zanzibar (Fig. 9). This result was further confirmed in 2008, where the CCA RDT was again tested using urine from 150 children on Unguja (Stothard et al., 2009c). Urine microscopy showed a mean prevalence of 30.7%, whereas CCA-positivity was only 4.0%.

Due to the lack of sensitivity of the CCA assays for *S. haematobium* infections, compared to the other *Schistosoma* species, there has been substantial focus on developing an antigen test that can be used also for urogenital schistosomiasis. Circulating anodic antigen (CAA) proved to be a suitable candidate as it was detectable for both *S. haematobium* and *S. mansoni* at high sensitivities (Corstjens et al., 2020). The first laboratory-based CAA assay was tested on 1,200 urine samples from children visiting schools in low prevalence areas on Pemba in 2013 (Knopp et al., 2015a). These urine samples, previously well-characterised by urine filtration and Hemastix reagent strips assessments, had been frozen as part of the sample repository associated with the ZEST project (Knopp et al., 2012). Two types of the up-converting phosphor-lateral flow circulating anodic antigen (UCP-LF CAA) assay (UCAA2000 and UCAA250) with the numbers representing the different volumes of urine concentrated for the tests, 2000 μL and 250 μL respectively, were performed in the well-equipped PHL-IdC on Pemba. The procedures for the test are detailed in Knopp et al. (2015a). In brief, a urine concentration step was performed using a centrifuge and custom-made CCA lateral flow strips were placed in a UCP-conjugate/sample mix. Subsequently, the strips were read using a portable strip reader. The procedure took about three hours but most strips were left over night before being read. The study revealed a *S. haematobium* prevalence of 13.9%, 4.7% and 4.1% for UCP-LF CAA, microscopy and microhaematuria respectively. Latent class analysis revealed a sensitivity of 97.0%, 85.5% and 66.7% for UCP-LF CAA, microscopy and microhaematuria respectively. It was concluded that the UCP-LF-CAA assay had a high diagnostic sensitivity for *S. haematobium* in low-endemic settings. However, limitations exist with the resources needed to run the assay, how long it takes, and the urine concentration steps needed. An RDT format for the CAA test would certainly support further testing in low endemicity settings such as Zanzibar (Corstjens et al., 2020).

8.7 Molecular (DNA) diagnostics

Molecular diagnostics show superiority in terms of their specificity and sensitivity, however the need for skilled personal, laboratory infrastructure, high costs and multiple steps often limits their use in endemic settings (Weerakoon et al., 2015). Three studies have investigated the use of molecular tests for diagnosing *S. haematobium* infections in Zanzibar, confirming that molecular testing is highly sensitive and specific but that major modifications are needed for implementation in endemic settings.

The studies also took advantage of the repository of characterised frozen ZEST urine samples on Pemba (Knopp et al., 2012).

The first study investigated a newly developed Dra1 qPCR assay in 2018 (Keller et al., 2020), which requires a fully functioning laboratory and skilled personnel. It showed that, when 792 frozen urines were tested and compared with urine filtration results a high sensitivity of 89.5% and a specificity of 82.3% was achieved. Sensitivity was very high (96.4–100%) when egg counts were reasonably high (10+ egg counts) but decreased with lower egg counts and was only 80.6% for ultra-low egg count samples (1–4 eggs/10 mL urine).

The second and third studies were based on an isothermal method called Recombinase Polymerase Amplification (RPA) (Rostron et al., 2019, Archer et al., 2020). This method was chosen due to its simplicity, speed, portability, and potential for becoming a future point of care test. Processing urine samples for molecular assays is simpler than processing faecal or blood samples making urine a preferred sample type for diagnostic developments. Additionally, with only urogenital schistosomiasis present in Zanzibar, diagnostic assessments are more straightforward. In a pilot study, 20 frozen urine samples were tested with the newly developed Sh-RPA assay giving positive results for urine samples with different eggs counts ranging from 1- > 500/10 mL of urine (Rostron et al., 2019). The method was robust and included a simple and low-resource sample preparation method. Even though RPA presents many advantages in terms of performance and feasibility for use in low resource settings, the current costs ($3.49–6.98 USD) of the assay and limited availability of the reagents are currently a limiting factor for large-scale use for schistosomiasis (Mesquita et al., 2022). However, the future use of a test such as RPA should not be ruled out as when the demand for novel tests increase, the associated costs tend to decrease. Also, despite increased cost, the highly accurate tests usually outweigh the perhaps cheaper but less accurate tests due to the economic cost of the disease (a consequence of missed infections) alongside the cost savings made from reduced treatment needs, resulting in the highly accurate tests being overall more cost-effective.

This encouraging pilot result led onto a larger study testing 200 urine samples (Archer et al., 2020). Small aliquots (50 μL) of each urine sample were processed using the rapid extraction method and tested using the Sh-RPA assay. Specificity of the Sh-RPA was 100% throughout, and 100% sensitivity was achieved for samples with very high egg counts (>400 eggs/10 mL urine) with 96.3% sensitivity achieved for samples with high egg counts

(50–399 eggs/10 mL urine). However, as for the qPCR and most other diagnostic assays, sensitivity of the Sh-RPA decreased with decreasing egg counts but even for samples with egg counts categorised as low (10–49 eggs/10 mL of urine) or ultra-low (1–9 eggs/10 mL of urine) intensity, a sensitivity of 94.2% and 91.4%, respectively, was achieved. It was concluded that with further development and optimisation, particularly for the sample preparation step, sensitivity could further increase.

Molecular (DNA) based diagnostics, although presenting logistical and cost limitations, certainly present the sensitivity and specificity levels needed for *S. haematobium* diagnosis in low endemic settings such as Zanzibar. With further developments, modifications and testing they have the potential to be used, in combination with other cheaper and low-cost screening tools, to provide accurate prevalence data and enable more targeted interventions, such as test-and-treat.

8.8 Outlook

Until recently, MDA with praziquantel without prior diagnosis of infection has been accepted as the 'go to method' for schistosomiasis control with a main aim of reducing associated morbidity. However, as prevalence and intensity of infections dramatically decrease in some endemic countries that are targeted by intense control and elimination efforts, such as Zanzibar, with obvious morbidity not often observed, more emphasis has been based on diagnostics. To move beyond control towards elimination and even interruption of transmission, accurate diagnostics that can identify asymptomatic infected individuals, who likely have low intensity infections, are needed. This will enable targeted timely treatment to prevent them contributing to transmission. Accurate diagnostics are also needed to aid decision making about treatment regimens and other intervention strategies as these rely on accurate infection data. There is also a need for diagnostics that can inform on treatment outcomes at the individual level, allowing follow-up treatment and also assessment of treatment efficacy (Hoekstra et al., 2021). Both CAA and DNA based diagnostics are probably the most promising platforms currently being used/developed for elimination settings. However, both come with their own limitations preventing their use as a true point of care test. Ongoing research will certainly move these platforms forward so that elimination is reached and maintained. Beyond prevalence and infection diagnosis, associated hidden morbidities, such as FGS (Bustinduy et al., 2022), need to be identified and treated in a timely manner before they become chronic and irreversible. It is clear that one

type or form of diagnostic is not going to fulfil all these criteria, with individual diagnostics having to fit each specific need. As with other diseases multiple diagnostic platforms should continue to be explored and evaluated.

Zanzibar presents a unique urogenital schistosomiasis endemic foci for diagnostic research, from understanding what kind of diagnostics are needed and feasible, to the evaluation of new diagnostic tests. Additionally, due to focused interventions, Zanzibar has experienced a rapid evolution in disease epidemiology (decline in prevalence and intensity of infections) providing a platform to evaluate the type of diagnostics that are applicable at the different stages of schistosomiasis control through to elimination. This review has highlighted several diagnostics that have been tested and deemed unsuitable, whilst others have provided valuable epidemiological data helping guide interventions to reach urogenital schistosomiasis elimination as a public health problem. Even the most basic diagnostics have proven to be highly informative in Zanzibar and truly shown their utility of guiding early interventions. As we move further forward in sustaining and going beyond elimination it is clear that gaps exist in the tools we have available to accurately determine infection prevalence. However, great scientific advances in diagnostic technologies have been made across the globe within the last decade and these are being capitalised to advance surveillance in Zanzibar and beyond.

9. Morbidity

Morbidity due to urogenital schistosomiasis has long been recognised in Zanzibar. Reports from the Medical, Sanitary and Biological Divisions of the Zanzibar Protectorate for the years 1929 to 1933 indicated that schistosomiasis was widely spread throughout Unguja and Pemba and was most commonly found in children and young people. However, at that time, it was reported that the disease appeared to be fairly mild and apparently became asymptomatic without any specific treatment (Zanzibar Protectorate, 1930–1934).

9.1 Radiological abnormalities discovered with urograms

The first scientific study that assessed the medical and public health importance of schistosomiasis was conducted in the Donge area on Unguja in 1964–1965 by Forsyth and Macdonald (1966). In this cross-sectional

survey including 517 schoolchildren, the *S. haematobium* prevalence was 45.3% and the urograms produced by intravenous pyelography showed pathology exclusively in children voiding *S. haematobium* eggs in their urine. A deformed ureter was found in more than 25.0% of *S. haematobium*-positive children, and a calcified bladder or hydronephrosis was detected in more than 10.0%. Initial lesions were more common in the younger children. Often, by the time of recruitment of children aged 6–13 years, ureteral deformation and calcification of the bladder had occurred already, hence, the beginnings of the pathological process could not be observed. Calcified bladders and uretic deformities were directly associated with the level of urinary egg excretion.

The *S. haematobium*-positive children from the study conducted in 1964–1965 were treated with monthly intramuscular injections of 10 mg/kg stibocaptate (Forsyth and Rashid, 1967b, Macdonald and Forsyth, 1968, Macdonald et al., 1968a). Two years later, in 1966, 149 among the formerly 234 positive children were re-examined for infection and urogenital pathology (Macdonald and Forsyth, 1968). While 43.3% of the children were *S. haematobium*-positive on this follow-up, the intensity of infection had decreased considerably and the mean egg output had dropped from 577.2 to 3.9 eggs. However, there was little difference in the prevalence of urological lesions in 1964/1965–1966, and no regression of bladder calcification was observed. Ureteric deformity and hydronephrosis had become less marked or could no longer be detected in some children, while in others it was seen for the first time. In general, in the longitudinal part of the study, the change in anatomy was not related to parasitological cure. *Schistosoma haematobium* infection and urogenital pathology were not associated with height, weight, anaemia, or school absenteeism.

In addition to the schoolchildren, the population of four contiguous hotspot villages (Fuasini, Karange, Mbiji and Mnyimbi) in the Donge area were investigated for urogenital schistosomiasis and related urogenital pathologies for two years, from 1964/1965–1966/1967 (Forsyth, 1969). Almost the whole community (1,004/1,074 inhabitants), including all ages, participated in the examinations for *S. haematobium* infection. Among them, 654 (56.1%) were voiding *S. haematobium* eggs. In children aged 7–17 years, prevalence was 90–100%. Regarding clinical manifestations, it was found that young children often had blood in urine but that this sign was less common in older children and adults. Five types of radiological abnormality were recorded, all of which were assumed to represent irreversible urological lesions: bladder calcification, ureteric deformity,

hydronephrosis, non-functioning kidney and urinary calculus. Among 794 participants with urograms, 281 (35.4%) had an abnormality, 111 (14.0%) had a calcified bladder, 189 (23.8%) had a deformed ureter, 119 (15.0%) had hydronephrosis, and 36 (4.5%) had a non-functioning kidney. All types of abnormalities were significantly more common in males than in females. The prevalence of bladder calcification showed an increasing trend from early childhood into later adult life, whence it decreased, potentially due to selective mortality caused by the association of bladder calcification with other pathologies, such as a non-functioning kidney. Ureteric deformity increased with age in children, dropped in adolescence, and became relatively static during adulthood. According to the authors, the reduction in ureteric deformation, as well as of hydronephroses, in adolescence might be explained by the spontaneous resolution of lesions due to reduced excretion of eggs. With increasing age, hydronephrosis and non-functioning kidneys became more common in adult men. Adults with hydronephrosis were more prone to develop a radiologically non-functioning kidney than those without hydronephrosis. Over the study period, 12.0% (3/25) of individuals with a non-functioning kidney died, compared with 3.3% (16/485) of adults that had a normal kidney, or only slight or moderate renal dysfunction. It was concluded that patients with a radiological non-functioning kidney due to a *S. haematobium* infection have a poor prognosis (Forsyth et al., 1970). After a second examination of the participants towards the end of the study period, the authors concluded that ureteric deformity and hydronephrosis caused by urogenital schistosomiasis are not always reversible, and were hence described as 'irreversible', with some persisting lesions that may progress towards renal failure and cause premature death (Macdonald et al., 1968a).

Cohen (1974) conducted an analysis in 1974 focusing on the economic benefits that would result from the elimination of mortality due to schistosomiasis and indicated that the study described by Forsyth (1969) needs to be interpreted with care. It neither offers a demonstration that a present or prior *S. haematobium* infection was associated with kidney failure among living participants, nor does it provide direct evidence that an infection with *S. haematobium* was the only or partial cause of pathology in the three deaths due to a non-functioning kidney (Cohen, 1974).

Based on the results of the studies reported above, a report from the WHO experts Mccullough & Krafft (1976) concluded that the public health importance of urogenital schistosomiasis in children in Zanzibar was considerable and deserved high priority in health promotion planning.

They estimated that 20% and 60% of the population on Unguja and Pemba, respectively, had or had had an infection with *S. haematobium* and that among those, 10% carried a 'heavy paired-worm load', which they regarded as a risk factor for 'serious vesical schistosomal disease', which was sometimes irreversible (Mccullough & Krafft, 1976).

9.2 Exceptional case report findings

Between the 1970s and the late 1980s, with the exception of several studies that assessed macro- and microhaematuria as a proxy for *S. haematobium* infection and related morbidity on Pemba (Savioli et al., 1989a, Savioli et al., 1989b, Savioli and Mott, 1989, Savioli et al., 1990b, Lwambo et al., 1997b), there was a paucity of larger studies focussing on the morbidity and public health importance of schistosomiasis in Zanzibar. However, two interesting case reports were published in 1984 and 1990, respectively. The first one documented a case of acute intestinal obstruction associated with *S. haematobium* infection in Pemba (Iozzi et al., 1984). The 18-year old male was referred from Vitongoji dispensary to Chake hospital with a history of abdominal pain and vomiting. At the hospital, the condition of the case worsened and surgery of the abdomen was performed to examine the abdominal organs (laparotomy). The appendix appeared to be long, bent and with a large tumefaction and had strangulated the terminal ileum resulting in intestinal obstruction. Once the appendix was removed and the patient had received antischistosomal treatment and additional care, he recovered to good health after 6 months. The histopathological examination of the removed appendix showed a thickened and fibrotic mucosa in which many *S. haematobium* eggs were found. It was concluded that the heavy ovideposition in the appendix had caused an intestinal strangulation that required emergency surgery. In addition, it was pointed out that the patient lived in a highly endemic area for *S. haematobium* on Pemba, and since only *S. haematobium* eggs were found in the appendix, the pathology and complication was with very high certainty attributable to *S. haematobium* (and not *S. mansoni*) infection.

The second case report documented the healing of a vulvar lesion caused by *S. haematobium* eggs with praziquantel treatment (Savioli et al., 1990a) and is hence the first report about an incidence of FGS in Zanzibar. A 9-year old girl from Pemba attended the Schistosomiasis Control Programme clinic in 1988. She presented with a painless lesion of the vulva that had been present for about four months and had gradually enlarged. In addition, she had two polypoid nodular growths of the left labium major

and hypertrophy of the left labium minor. Gently scraping of one of the growths with the corner of a glass slide, immersing the scraping in saline, and examining under a microscope revealed many *S. haematobium* eggs mixed with tissue debris. Urine filtration and reagent strips showed a heavy intensity infection and heavy microhaematuria (+++). The girl was treated with praziquantel (40 mg/kg) and one month later, the nodular lesions were greatly reduced. Six months later, the nodular lesions, the hypertrophy of the labium minor and the haematuria had resolved. Savioli et al. (1990a) indicated that in areas where *Schistosoma* spp. are endemic, routine biopsies of proliferative genital lesions had been recommended to distinguish them from lesions caused by sexually transmitted infections (Attili et al., 1983). Moreover, before praziquantel became available, the management of vulvar schistosomiasis was surgical excision followed by chemotherapy (Janovski and Douglas, 1972). Based on the observations from the case of the 9-year old girl from Pemba and since histological facilities are mostly not available in small rural hospitals in Africa, Savioli et al. (1990a) suggested to examine lesions by simple gentle scraping and subsequent microscopy before an invasive procedure is conducted and also recommended to treat cases with praziquantel and to wait six months before any other possible measures are taken.

9.3 Uropathy discovered with ultrasonography

In 1988, Hatz et al. (1990) conducted a cross-sectional study in Pujini, a highly endemic village on Pemba. Participants of all ages were examined by ultrasonography for uropathy, with Hemastix reagent strips for microhaematuria and with urine filtration for *S. haematobium* eggs. Compared with males, females showed a considerably lower uropathy, despite a similarly high prevalence of microhaematuria and *S. haematobium* infection. The authors suggested lower infection intensities and an underlying lower exposure in females as a potential explanation. While they highlighted that their methodological approaches were different to the study conducted by Forsyth (1969) and results therefore not directly comparable, they attributed lower rates of kidney pathological signs found in 6–14 year old children on Pemba to the new treatment possibilities that had been developed over the past 20 years and treatment of younger age groups by the Schistosomiasis Control Team on Pemba. Of note, two MSc projects were conducted by students from the London School of Hygiene and Tropical Medicine in 2000 and 2010, respectively, which also assessed uropathy, microhaematuria and *S. haematobium* infection in Pujini's

population (Deganello, 2000, Groenendael et al., 2010). While the prevalence of microhaematuria and *S. haematobium* infection was very similar in the studies conducted in 1988 and 2000, there was a significant drop in the microhaematuria prevalence from 2000 to 2010 (Savioli et al., 1990b, Deganello, 2000, Groenendael et al., 2010). The levels of uropathy had not much changed (Hatz et al., 1990, Groenendael et al., 2010). Since no adjustments for age and sex were made, the results need to be interpreted with care, however.

Urogenital schistosomiasis and urogenital tract pathologies were also assessed in a study conducted in the Chaani area on Unguja in 2005, including randomly selected males attending one health facility and schoolchildren in three schools in 2005 (Sousa-Figueiredo et al., 2009). The *S. haematobium* prevalence was 46.8% in adult males (19.2% heavily infected) and 36.4% in schoolchildren (15.7% heavily infected). Urogenital tract pathologies were found in 64.4% of men and in 39.4% of schoolchildren. Bladder pathologies, primarily bladder wall irregularities, were most commonly observed. In children, active *S. haematobium* infection was associated with pain on urination and with urogenital tract pathologies, particularly with bladder or ureteral pathologies. In adult males, urogenital schistosomiasis was associated with pain on urination and urine flow problems, reflecting the damage caused by egg expulsion through the urogenital tract. However, *S. haematobium* infection was not associated with urogenital tract pathology in general nor with any individual pathologies of the bladder, ureter or renal pelvis.

In another study conducted one year later in 2006, the use of the urine albumin-to-creatinine ratio (UACR) to predict *S. haematobium* infection and urogenital tract pathology, respectively, was evaluated in 62 schoolchildren living in a highly endemic area for *S. haematobium* on Unguja (Stothard et al., 2009b). Many children were infected with *S. haematobium* (65.2%), had heavy intensity infections (33.3%), microhaematuria (77.3%) and urogenital tract pathologies (66.1%), and according to the authors urogenital tract pathology was highly associated with *S. haematobium* infection in the population studied (but data were not shown). Abnormal (≥3.4 mg/mmol) and severely abnormal (≥33.9 mg/mmol) UACRs were highly sensitive indicators for detecting active infections and/or urogenital tract pathology, however had only low to moderate specificity.

In 2007, Lyons et al. (2009) assessed the prevalence of *S. haematobium* infection, urogenital tract pathologies and self-reported symptoms in a study involving 160 people aged ≥16 years from high endemic areas

(Chaani, Mwera, Kinyasini and Upenja) and 101 individuals from low endemic areas (Paje and Muyuni) in the north and south of Unguja, respectively. Compared with individuals living in the south of Unguja, participants from the high endemic area in the north were much more often infected with *S. haematobium* (10.2% versus 0%), microhaematuria-positive (8.3% versus 0%) and showed urogenital tract pathology (17.3% versus 1.0%) (Lyons et al., 2009). Predictors for urogenital tract pathologies, diagnosed with ultrasonography, were self-reported urogenital schistosomiasis in the past year, self-reported urgency to urinate and *S. haematobium* eggs identified in urine.

9.4 Anthropometric measures and anaemia

A study related to morbidity factors associated with urogenital schistosomiasis was conducted on Pemba in 1994. Here, Stoltzfus et al. (1997a), assessed the cross-sectional and longitudinal patterns of growth in more than 1,000 primary schoolchildren from four schools on Pemba over one year. Anthropometric measurements of weight and height were taken, and besides anaemia, *Plasmodium*- and soil-transmitted helminth infections were assessed. The presence of macro- or microhaematuria was used as an indirect proxy for *S. haematobium* infection. Significant linear growth retardation was observed in children during their primary school years. A marginally significant association between microhaematuria and poor linear growth increment was identified, but the authors pointed out that the observations were statistically not very strong and explained only a small fraction of the variability in growth.

In the cross-sectional baseline survey of the longitudinal study highlighted above, the authors also collected data on iron deficiency anaemia from 3,595 children from 12 schools across Pemba (Stoltzfus et al., 1997b). However, no association between haematuria-positivity and iron deficiency anaemia was detected. In addition to haematuria being an imperfect diagnostic indicator for schistosomiasis, the effective test-and-treat campaign that was conducted from 1988 to 1992 across Pemba (see section Treatment interventions), and the consequently rather short duration of *S. haematobium* infections in children participating in the study, were suggested as potential reasons for the lack of association in this study. However, earlier and later studies conducted on Unguja also did not show an association between *S. haematobium* infection and anaemia in the populations studied (Forsyth, 1969, Forsyth, 1970, Sousa-Figueiredo et al., 2008, Knopp et al., 2010), and hookworm infection, stunting, or self-

reported malaria over the past two weeks were stronger explanatory variables (Stoltzfus et al., 1997b, Knopp et al., 2010).

In addition to anaemia, growth retardation was assessed in detail in a study focusing on urogenital schistosomiasis in preschool-aged children and their mothers conducted in the area of Chaani on Unguja in 2006 by Sousa-Figueiredo et al. (2008). All the mothers who volunteered to participate (113) and their children aged ≤6 years (152) were enrolled. *Schistosoma haematobium* eggs were detected in the urine samples from 12.7% of mothers and 3.9% of children. Anaemia was very common in children (73.4%) but less so in mothers (25.9%). Chronic, acute and general under-nutrition were found in 38.5%, 14.7% and 2.2%, respectively, of the children, but the nutritional indices and mid-upper-arm-circumference were not associated with a *S. haematobium* infection, nor with *Plasmodium*- or soil-transmitted helminth infections.

9.5 Outlook

Over the past decade, again, there has been a paucity of studies focussing on morbidity due to urogenital schistosomiasis in Zanzibar. However, over the long period of control and elimination interventions that started in the 1980s and remain ongoing, *S. haematobium* prevalence, infection intensities, microhaematuria and, therefore, likely also acute morbidity, were substantially reduced (Knopp et al., 2019b, Trippler et al., 2021a). Anecdotally, there is evidence that urogenital tract pathology is less common nowadays and, hence, it has been concluded that the MDA programmes in Zanzibar have successfully reduced the burden of disease in the population (Ali et al., 2015). According to the WHO criteria from 2020 (World Health Organization, 2020), Zanzibar achieved elimination of schistosomiasis as a public health problem in 2017 (Knopp et al., 2019b, Knopp et al., 2019c, Trippler et al., 2021a). However, little to nothing is known about the chronic sequelae in a population living in a formerly highly endemic area, nor about persistent morbidity signs in the remaining hotspot areas, and about the extent of FGS in Zanzibar to date. For the latter, a qualitative study has been conducted, which showed that while most people on the Zanzibar islands were aware of urogenital schistosomiasis, they lacked knowledge about FGS and were not aware that an infection with *S. haematobium* can affect the female reproductive system (Mazigo et al., 2021). However, with the exception of the case report of the vulvar lesions described above (Savioli et al., 1990a) the existence and extent of FGS in Zanzibar has not yet been reported and assessed.

Hence, there is a need for future research that shows that Zanzibar has indeed eliminated urogenital schistosomiasis as a public health problem, not only according to the definitions currently provided by the WHO, but also considering and assessing morbidity potentially caused by light intensity infections (as those are most prominent in Zanzibar and other elimination settings), due to FGS, and also in terms of urogenital tract pathologies that may be associated with a present or past *S. haematobium* infection.

10. Treatment interventions

10.1 Early antischistosomal treatments in Zanzibar

A very early indication of schistosomiasis treatment on the Zanzibar islands in written literature can be found in reports from the 'Medical, Sanitary and Biological Divisions of the Zanzibar Protectorate for the year 1929–1933' (Semple, 1927, Zanzibar Protectorate, 1930-1934). Semple (1927) highlighted that Zanzibar residents were seeking treatment for symptoms likely associated with schistosomiasis at government hospitals and dispensaries. Treatment of schistosomiasis during this time would have consisted of at least one injection with a substance that although not named in the reports from Zanzibar, likely consisted of toxic chemicals such as tartar emetic (Cioli et al., 1995). It is indicated that many individuals treated in Zanzibar did not return for additional injections after the immediate symptoms were relieved, perhaps because of the unpleasant side effects of the treatment (Semple, 1927). A later report from 1929 described two cases where antimosan or neo stibosan were injected intramuscularly (Zanzibar Protectorate, 1930–1934). Antimosan resulted in a rapid disappearance of all symptoms, and although insufficient time elapsed to judge whether a permanent cure had occurred, the physician in charge recommended its use in cases where intravenous injections were difficult due to the absence of large superficial veins. This might imply that other antischistosomal drugs used at that time were injected intravenously.

Four decades later, during the 1960s, Unguja became the site of a series of studies to assess the efficacy, safety and tolerability of several promising antischistosomal drug candidates and their most favourable dosage and administration regimen for large-scale interventions.

Forsyth (1965) conducted an experiment testing several regimens of antischistosomal drugs in children from Donge school, which showed that monthly injections of stibocaptate (10 mg/kg) (also known as astiban) were

the most favourable treatment option in terms of tolerability, cure rate and convenience among schoolboys infected with *S. haematobium*. Recognising the challenges of a treatment administered through intramuscular injections, six studies were conducted between 1965 and 1967 in schools in the shehias Donge, Mfenesini, Kinyasini and Chaani assessing drugs that could be taken orally (Forsyth, 1966, Forsyth and Rashid, 1967b, Macdonald et al., 1968b). These drugs included niridazole (also known by the trade name Ambilhar), lucanthone hydrochloride (nilodin or miracil D) or trichlorophone (metrifonate). All treatments reduced the excretion of *S. haematobium* eggs in the schoolchildren and no toxic side-effects were reported (Forsyth, 1966, Forsyth and Rashid, 1967b, Forsyth and Rashid, 1967a, Macdonald et al., 1968b). It was concluded that repeated single oral doses of niridazole or trichlorophone might be suitable for large-scale treatment schemes due to their ease of administration (Forsyth, 1966, Forsyth and Rashid, 1967b, Macdonald et al., 1968b). Furthermore, the researchers aimed at estimating and quantifying the morbidity arising from urogenital schistosomiasis, as well as the effects antischistosomal treatment had on the clinical signs and symptoms caused by the disease (Forsyth and Macdonald, 1966, Macdonald and Forsyth, 1968, Macdonald et al., 1968a, Forsyth, 1969). To this end, a longitudinal study was conducted between 1964 and 1966, which involved the clinical and parasitological examination at two time points with intermediate treatment of schoolchildren from Donge school (Macdonald and Forsyth, 1968). Schoolchildren who tested positive for *S. haematobium* by urine filtration were given monthly injections of stibocaptate (10 mg/kg) until they were found to be egg-negative; leading to the conclusion that the prevalence of urological abnormalities in schoolchildren could be substantially reduced by monthly stibocaptate injections (Macdonald and Forsyth, 1968, Macdonald et al., 1968a). These various treatment efforts were also combined with molluscicide (to reduce the snail host population) of water bodies identified as possible transmission sites. These combined intervention efforts reportedly increased the effectiveness of the treatment campaigns in reducing transmission.

Between 1965 and 1967, a pilot mass-treatment programme was conducted, again in Donge school, and the surrounding communities with the aim of 'eradicating' *S. haematobium* infections (Forsyth, 1966, Macdonald et al., 1968b, Forsyth, 1969). Notably, the author's use of the term 'eradication' should not be considered equivalent with today's epidemiological definition but is rather used as a synonym for cure at the individual level and for disease control at the population level (Dowdle, 1998). A mass-treatment

programme was implemented where infected community members and schoolchildren were treated with monthly doses of orally administered niridazole or lucanthone hydrochloride, and also occasionally injections of stibocaptate until participants presented as egg-negative by urine examination or were lost to follow-up between September 1965 and December 1966 (Forsyth, 1966, Macdonald et al., 1968b). Additionally, water bodies identified as transmission sites were systematically surveyed for snails and treated with molluscicide if *Bulinus* were found. Although this pilot project did not result in the interruption of transmission or cure of all infections in the study population, three key lessons were learnt, which were documented by Macdonald et al. (1968b) and have ultimately helped shape future treatment programmes in Zanzibar. First, the success of any 'eradication' campaign lies on the cooperation of local authorities and communities. Second, the oral administration of single-dose drugs was reported to be well accepted, however repeated administrations were necessary for cure and most participants were lost to follow-up after three doses, highlighting that the drug of choice needs to be efficacious within one or two doses. Third, a door-to-door distribution of drugs was concluded to be an optimal approach for community-based treatment.

10.2 Early test-and-treat approaches and community-wide selective chemotherapy

Beginning in 1975, the government of the Zanzibar islands, and WHO officers and consultants initiated concrete plans to set up a national control programme for schistosomiasis (Manning, 1976, Mccullough & Krafft, 1976, WHO Archives, n.d., Mccullough & Krafft, 1976) from the WHO, who collaborated with Zanzibar authorities, noted that control efforts had so far relied on chemotherapy administered to patients seeking treatment at hospitals and primary healthcare clinics. The drug of choice at this time was niridazole (Ambilhar), which was comparatively costly and not available in sufficient quantities to treat the entire affected population [estimated at 20% on Unguja and a staggering 60% on Pemba (WHO Archives, n.d.)]. Furthermore, niridazole (Ambilhar) did not result in satisfactory cure rates if the full course of more than three doses was not adhered to, which the authors stated as a likely reason for persisting high rates of infections despite treatment being implemented (Mccullough & Krafft, 1976). Therefore, the use of metrifonate (trichlorfon), which was much cheaper and reported to lead to very few side-effects, was suggested. Nearly a decade later, building on the successes and conclusions drawn by Macdonald et al. (1968b),

an ambitious and pioneering approach was drafted, involving a pilot intervention phase in at least two highly endemic areas with expansion to all areas with a *S. haematobium* prevalence ≥20% among 6–16 year old male children in a second phase (Mccullough & Krafft, 1976). It was suggested that molluscicide of water bodies should be applied together with health education in addition to treatment (see sections Snail control interventions and Behaviour change interventions). The treatment plan was targeting school-aged children (6–18 years) according to the following eligibility criteria: treating all children and young adults in areas with a prevalence of over 50%, applying a test-and-treat strategy for children and young adults in areas of prevalence between 20% and up to 50%, and referring positive cases in areas with below 20% prevalence to hospitals and health clinics for treatment. The treatment regime was to consist of three doses at fortnightly intervals and administered in two courses, with the second round to take place one year after the first treatment. The original plans were re-discussed and refined until 1978 (Manning, 1976, Mccullough, 1978). However, in 1979, Mccullough et al. (1979) referred back to the schistosomiasis control activities suggested by Mccullough & Krafft (1976) and stated that although the 'initiation of these activities is commendable', treatment had been given only unsystematically and mostly without evaluation afterwards. As a result, no satisfactory benefits were expected.

Implementation of broader scale interventions finally occurred in the 1980s, when two multi-year control efforts involving entire communities were conducted with systematically measured outcomes, which included the reduction in prevalence and intensity of *S. haematobium* infections (Savioli et al., 1989b, Mgeni et al., 1990). The first study was conducted in Kinyasini shehia in the north of Unguja between 1981 and 1983 by officers of the Zanzibar MoH, with the support of the WHO (Mgeni, 1984a, Mgeni et al., 1990). The entire population of the shehia was invited to provide urine samples, which were tested for *S. haematobium* by urine filtration. Subsequently, participants that were egg-positive were treated with three doses of metrifonate at 2-week intervals, which was the regime recommended by the WHO at that time and also previously suggested by McCullough and Krafft (Mccullough & Krafft, 1976, Mgeni, 1984a, World Health Organization, 1985). A total of four surveys were conducted over the 2-year period, in which the entire population present in the district was invited for testing and offered treatment if tested positive (Mgeni et al., 1990). Additional interventions including health education and information sessions at Kinyasini school, efforts to improve the supply of safe water,

and molluscicide of the main transmission sites were also implemented (see sections Snail control interventions and Behaviour change interventions). The study met its target of reducing the *S. haematobium* prevalence by at least 50% with an overall reduction of 52.9%. However, heavy infections (≥50 eggs per 10 mL of urine) were only reduced by 62.2% as opposed to the targeted 75%.

Three years later, in 1986, an island-wide test-and-treat control programme was initiated on Pemba under the direction of the WHO and supported by the Zanzibar MoH and the Italian Agency for Development Cooperation, and with funding from the German Pharma Health Fund from 1987 onwards (Muchiri, 1991, Savioli, 1991a, Lwambo et al., 1997b). The programme ran for five years and aimed to eliminate clinical morbidity arising from *S. haematobium* infections. Urine samples were assessed for macro- and microhaematuria using Hemastix reagent strips, which served as a morbidity marker and as an indicator for surveillance needs, including the identification of individuals for targeted selective chemotherapy (see section Diagnostics). Individuals that tested haematuria-positive were treated with a single dose of praziquantel (40 mg/kg) (Savioli et al., 1989b). In 1984, praziquantel had been recommended for the treatment of schistosomiasis by the WHO and went on to become the drug of choice for large-scale treatment schemes (World Health Organization, 1985). Schoolchildren were tested with Hemastix reagent strips and treated eight times at 6-month intervals, whereas the entire population received the test-and-treat intervention two times during the five years (Savioli, 1991a). By the end of the programme in 1990, an overall reduction of haematuria, both macro- and microhaematuria, to <10% and a reduction of heavy haematuria to <0.5% were achieved (Savioli, 1991a). Muchiri (1991) reported that the test-and-treat method was well accepted by the schoolchildren and the communities due to several factors, such as the culturally appropriate implementation of the testing, including offering two separate queues for male and female participants to provide urine, and the Hemastix reagent strips showing an instant test result (providing point of care diagnostics) (Muchiri, 1991). Furthermore, the assessment with Hemastix reagent strips was reported as being an adequate community-wide indicator of prevalence and heavy intensity infection and thus, as a proxy for morbidity (Savioli et al., 1989b, Savioli, 1991a, Lwambo et al., 1997b, Chan et al., 1999). There is evidence that further test-and-treat campaigns within the 'Schistosomiasis Manual' were planned in the 1990s (Mgeni, 1995a), and although there are records alluding that they were implemented

afterwards (Evans, 1997, Mgeni, 2002) there was no documented evaluation of the test-and-treat methods executed.

These early test-and-treat programmes conducted in the 1980s laid the foundation for studies that started decades later, such as a test-and-treat intervention that has been implemented as part of the currently ongoing SchistoBreak project for urogenital schistosomiasis elimination in the north of Pemba from 2020 to 2024 (Trippler et al., 2021b). In SchistoBreak, in shehias with low *S. haematobium* prevalence, a (re)active test–treat–track–test–treat (5T) approach is followed, where schoolchildren and community members are tested for urogenital schistosomiasis using Hemastix reagent strips and urine filtration, and treated with praziquantel if positive (see section Surveillance-response interventions).

10.3 National Helminth Control Programme: Preventive chemotherapy for morbidity control

On the basis of the success of the preceding 5-year programme from 1986 to 1990 (Savioli, 1991a), the Zanzibar MoH (then Ministry of Health and Social Welfare) went on to develop a national plan of action for the control of helminth infections in the early 1990s (Knopp et al., 2013b). Large-scale preventive chemotherapy programmes using praziquantel to treat schistosomiasis and albendazole to treat soil-transmitted helminths were set up in 1994 through the MoH's established National Helminth Control Programme (Alawy, 1995, Mgeni, 1995b, Mohammed et al., 2008, Knopp et al., 2012). These MDA campaigns were mainly school-based, with schoolchildren being the primary population targeted for treatment. Occasionally the treatment campaigns were also extended to the community (Mohammed et al., 2008). These MDA campaigns ran consistently for five years, with drug distribution becoming irregular between the years 1999 and 2003 and an interruption in delivery from 2000 to 2001 due to issues with drug procurement (Mohammed et al., 2008, Knopp et al., 2013b, He and Kabole, 2021).

In their 54th meeting in 2001, the World Health Assembly (2001) declared the target of deworming at least 75% of school-aged children by 2010 in endemic areas, which represented an important milestone for preventive chemotherapy schemes. Zanzibar was the location of choice for two studies under the lead of the WHO, examining extensions to the current methodologies of MDA programmes in the early years of the new millennium (Montresor et al., 2001, Montresor et al., 2002b). The first study, conducted in January and February 2000 in eight shehias in northern

and central Unguja, explored the possibility of extending anthelmintic drug coverage to non-attending school-aged children providing guidance for control programme managers (Montresor et al., 2001). By pre-announcing the treatment day to enrolled children in schools and extending the invitation to their non-school-attending but school-aged relatives and friends, a remarkable 60% of the non-school-attenders living in the eight shehias were able to be reached by treatment. The second study, conducted in April 2001, was designed to test the application of a dose pole (Fig. 10) in comparison to the formerly used weight scale for an easier administration of an appropriate dosage of praziquantel in the field (Montresor et al., 2002b).

Fig. 10 The praziquantel dose pole being used to quantify the number of tablets to treat a student in Zanzibar (personal collection, Stefanie Knopp).

After its validation, the dose pole went on to become a standard tool in deworming programmes recommended by the WHO (Montresor et al., 2002a) and remains in widespread use in Zanzibar and elsewhere (Montresor et al., 2002b, Montresor et al., 2005, Knopp et al., 2012, Sousa-Figueiredo et al., 2012, Gazzinelli-Guimaraes et al., 2018, Sakho et al., 2021, Trippler et al., 2021b).

The establishment of the national plan marked the beginning of MDA schemes in both Unguja and Pemba, making Zanzibar a pioneer of large-scale preventive chemotherapy programmes, which have been the cornerstone of global helminth control to date.

10.4 Establishment of a national schistosomiasis control programme and integrated MDA

In the wake of the increased attention given to deworming as the mainstay of endeavours of curbing morbidity and the disease burden on endemic populations, several countries established national schistosomiasis control programmes in the early 2000s (Savioli et al., 2004, Southgate et al., 2005, Scientific Working Group On Schistosomiasis, 2006, Tschuem Tchuenté, N'goran, 2009). On Unguja and Pemba, the *Piga Vita Kichocho* (Eng.: Kick Out Schistosomiasis) campaign was launched in 2003, run by the Zanzibar MoH with support from the NHM and the SCI (Stothard et al., 2009a, Knopp et al., 2013b, He and Kabole, 2021). Within the programme, praziquantel and albendazole were distributed annually to school-aged children on Unguja and to entire communities on Pemba (Stothard et al., 2009a, Guidi et al., 2010, Knopp et al., 2013b). Monitoring and evaluation surveys in 24 sentinel site schools on Unguja revealed a drop in the overall prevalence to below 10% by 2006 (Stothard et al., 2009a, Knopp et al., 2012).

In parallel to deworming efforts targeting schistosomiasis and soil-transmitted helminths, the MoH's Lymphatic Filariasis Elimination Programme, with support from the Liverpool School of Tropical Medicine, carried out annual MDA with ivermectin and albendazole on both islands between the years 2001 and 2006, treating everyone eligible above five years of age (Knopp et al., 2013b, Rebollo et al., 2015, He and Kabole, 2021). Treatment for lymphatic filariasis was coordinated with treatment for schistosomiasis and soil-transmitted helminths. A pilot study in one high-transmission setting in both Unguja and Pemba was conducted to integrate the MDA programmes against lymphatic filariasis, schistosomiasis and soil-transmitted helminths using triple co-administration of ivermectin, praziquantel and albendazole, respectively. No serious adverse

events were observed, leading to a national roll-out of this triple MDA approach at the end of 2006. After this triple therapy approach, the Lymphatic Filariasis Elimination Programme and the National Helminth Control Programme were combined and became the newly formed MoH's NTD Programme (Mohammed et al., 2008, Fenwick et al., 2021).

Two years later, in 2008, a cross-sectional study conducted in two shehias on Unguja (Bandamaji and Dole) concluded that the chemotherapy-based control efforts in Zanzibar could be considered a successful public health intervention due to the significant reduction of prevalence and infection intensities together with the high levels of treatment coverage achieved (Knopp et al., 2010). Nevertheless, the limitations of solely MDA-based schistosomiasis control were reflected in the persisting infection levels in specific foci with continued transmission (Knopp et al., 2010, Kim et al., 2020).

10.5 Aiming for elimination of urogenital schistosomiasis: The national plan and moving beyond MDA

The government of Zanzibar committed to the elimination of urogenital schistosomiasis in both Unguja and Pemba and established the national plan of Combatting NTDs in the years 2009 and 2010 (Knopp et al., 2012). The Zanzibar MoH was supported in this endeavour by the international ZEST alliance (see Introduction), and in terms of treatment efforts specifically by the WHO, which supplied praziquantel and by the SCI, which provided technical and financial support for the large-scale drug administration (Knopp et al., 2012).

During the ZEST project, biannual MDA with praziquantel for schistosomiasis together with albendazole and ivermectin for STHs and lymphatic filariasis, respectively, were distributed to the whole population of Unguja and Pemba (Ali et al., 2015, Knopp et al., 2019b). In 2014, three years into the ZEST study, a survey was conducted in 93 schools and 92 communities on Unguja and Pemba, aiming to evaluate the coverage of praziquantel treatment during the most recent MDA, which had taken place 3–4 months prior to the survey (Knopp et al., 2016). The results revealed a coverage of 86.9% in schoolchildren on Unguja and 85.2% on Pemba, while adult community members reported a coverage of 64.9% on Unguja and 53.6% on Pemba.

Over four years of implementation, from 2013 to 2016, a total of 2.4 million treatments were provided on Pemba with estimated implementation costs of 0.21 US$ per treatment, excluding the costs of donated

praziquantel (Salari et al., 2020). On Unguja, the same or a larger amount of treatments was provided, but no cost-related data are available. The cluster-randomised trial revealed that biannual MDA alone was not sufficient to completely interrupt *S. haematobium* transmission, and additional interventions of either snail control or behaviour change did not significantly boost the effect of MDA either (see section Epidemiology of schistosomiasis in Zanzibar) (Knopp et al., 2019c). It was also shown that as elimination approaches, the identification and treatment of infected individuals becomes more difficult due to very low egg outputs together with the higher chances of single sex schistosome infection (Hoover et al., 2020). However, it should be noted that in 2014, Zanzibar was included in a multi-country study to validate the therapeutic efficacy of praziquantel, which found that a single 40 mg/kg treatment with praziquantel resulted in a 99% egg reduction rate in the 90 individual cases that were examined (Levecke et al., 2020). Furthermore, in 2017, as part of a 'project to assist in the prevention and treatment of schistosomiasis in Zanzibar' (Zhou et al., 2019), a study was conducted in three shehias on Pemba involving 152 individuals, comparing the effectiveness of praziquantel from two different manufacturers: Merck KGaA in Germany, and Nanjing Pharmaceutical Factory Co Ltd in China (Wang et al., 2019, Yang et al., 2019). The study showed that both drugs were highly effective, regardless of their manufacturer.

After the ZEST project was concluded in 2017, MDA continued annually under the leadership of the Zanzibar NTD Programme until 2020 with a 16-months treatment gap between April 2019 and August 2020 due to drug procurement issues (Trippler et al., 2021a). Continued cross-sectional surveys revealed a prominent recrudescence of infection in schools with a history of moderate prevalence after the 16-month treatment gap in 2019/2020, highlighting the importance of continued high-quality MDA in areas at high risk, to maintain the successes achieved by control efforts over the last decades (Trippler et al., 2021a).

In addition to procurement issues, the COVID-19 pandemic caused an interruption to the cross-sectional school and community-surveys on Unguja and Pemba and a characteristics survey of shehias in northern Pemba participating in the 4-year 'SchistoBreak' research project (2020–2024) in 2020 (Trippler et al., 2021a, Trippler et al., 2022b). As a result, only a limited number of schools and shehias were surveyed in 2020 (67 schools and 68 shehias, instead of 90 and 90, respectively), and the characteristics survey was delayed until 2021. However, it should be noted

that there were fewer COVID-19 related restrictions in Zanzibar than there were elsewhere in sub-Saharan Africa, and hence, MDA to the population was not impacted.

The ongoing 'SchistoBreak' research project aims to investigate the field applicability and performance of a multidisciplinary intervention approach, with different interventions being implemented in shehias classified as either a low prevalence or hotspot area (Trippler et al., 2021b). Areas where the risk of *S. haematobium* transmission is high are targeted with biannual school-based and community-based MDA with praziquantel, snail control and behaviour change interventions (Trippler et al., 2021b, Trippler et al., 2022a). Areas with low risk for *S. haematobium* transmission receive targeted surveillance-response interventions, including a test–treat–track–test–treat approach with praziquantel (see section Surveillance-response interventions). The approach is in line with recent considerations of overtreatment, careful use of available praziquantel and other resources, and the community's desire for a diagnosis for schistosomiasis before antischistosomal treatment (Trippler et al., 2022b).

10.6 Outlook

With the knowledge gained since the national plan was first drafted in 2009 and in the light of both the WHO targets in the 2030 NTD roadmap and in the updated recommendations on schistosomiasis treatment (World Health Organization, 2020, World Health Organization, 2022), the Zanzibar MoH and its NTD programme continue their efforts to implement praziquantel MDA with a high coverage and programmatic reach. However, previous schistosomiasis research on the islands regarding treatment highlights various challenges that need to be addressed, such as the insufficient coverage and hesitancy among adults to participate in the MDA programme (Knopp et al., 2016, Trippler et al., 2022b). This hesitancy derives primarily from apparently healthy individuals who wish to be tested before being treated and because the drug distributors are not perceived as medical personnel (Trippler et al., 2022b). Hesitancy towards praziquantel MDA is not a problem unique to Zanzibar, being also observed in Uganda, where perceptions of the disease have led to a proportion of the population rejecting praziquantel treatment in favour of an herbal treatment from traditional healers, or rejecting praziquantel due to perceived side-effects (Mujumbusi et al., 2023). The integration of behaviour change interventions that increase awareness of schistosomiasis, including better knowledge about the transmission cycle and health consequences associated with

infection, and to emphasise the benefits of regular praziquantel treatments improve compliance during MDA, but they need to be implemented at large-scale and frequently to reach all people. The focality of *S. haematobium* transmission across the islands results in differing treatment needs in the low prevalence versus high transmission areas. While continuing MDA with a high coverage and compliance will be of utmost importance in areas with high transmission potential to further reduce prevalence and prevent morbidity, continuing MDA in low prevalence areas, which of note are the majority in Zanzibar, might lead to an overtreatment of a predominantly healthy population. For the low prevalence areas, a key step will be the development of a more refined and targeted treatment approach that is able to maintain the gains made or accelerate elimination. This may include the treatment of symptomatic patients in health facilities, including pregnant women and infants (Sousa-Figueiredo et al., 2011, De Rosa et al., 2022), risk-based test-and-treat approaches, or focal preventive chemotherapy to schools and households located near water bodies that are a habitat for intermediate host snails (Trippler et al., 2022b). In the context of targeted treatment, the development and implementation of highly sensitive and specific diagnostics that can be performed in a high throughput manner is essential to accurately identify all infected individuals. Although treatment alone may not be sufficient to reach interruption of transmission across Zanzibar, it will remain a backbone of Zanzibar's ongoing elimination strategy. It is crucial to maintain and build upon the great progress of the past achieved through extensive MDA programmes, to ensure that the impact of schistosomiasis on the health and well-being of the Zanzibari population remains low and progresses towards zero.

11. Snail control interventions

11.1 Snail control interventions in the 20th century

To date, snail control has involved the use of chemical molluscicides or the implementation of biological or environmental measures to reduce the number of intermediate host snails, the ultimate aim being to break the human-snail-human transmission cycle of *Schistosoma* (World Health Organization, 2017). The first person describing snail control in the context of schistosomiasis interventions in Zanzibar was Mansfield-Aders (1928). Besides treatment of humans, he suggested to destroy the intermediate host snails using chemicals such as copper sulphate and/or by

modifying their freshwater habitats to make them less favourable for snail habitation, for example by clearing swamps of plants. He also suggested encouraging residents to keep ducks as they were known to feed on the snails. In 1929, the drainage of a pool was considered that was found to be the source of many human schistosomiasis infections (Zanzibar Protectorate, 1930–1934). However, these measures were postponed due to a lack of resources.

A decade later, Mozley (1939) highlighted the need to find organic snail control substances derived from native plants due to preconceptions of Zanzibar's population towards the addition of 'unknown substances', such as chemical treatment, to their freshwater ponds. Therefore, in a laboratory-based study, he investigated the vitality of *B. globosus* after exposure to tap water from Zanzibar enriched with chopped material of different native plants. Although some of the plant material, such as the leaves and stems from *Psiadia arabica* 'Form 3' or the fleshy part of the fruit from *Sapindus saponaria,* seemed to have successfully killed the snails, the experiment was not extended to a field study in open freshwater bodies on the Zanzibar islands. Hence, the impact of respective Zanzibar native plant material on natural populations of *Bulinus* remains unknown. It was also Mozley (1939), who suggested the construction of a drainage ditch controlled by a removable dam at a large marshy pond in Muyuni in southern Unguja, where a high infection rate of *S. haematobium* had been found in the area. Mozley singled out Muyuni pond because *Bulinus* were found in great abundance and it was routinely used for bathing, conducting domestic chores and for providing a water source for rice irrigation. Since the landscape also proved suitable for constructing an effective drainage ditch, it was then constructed shortly after (Fig. 11). Closing the dam during the dry season and draining the pond by a ditch was done to reduce the number of snails, whilst opening of the dam during the rainy season allowed the rice to fully develop and thus ensure the agricultural yield of the pond. Observations from Stothard et al. (2002d) in 2001 showed that the vegetation at the Muyuni pond had clearly changed from before the ditch was built and that intermediate host snails were no longer present (Fig. 11). The pond is evidently no longer permanent, with a site visit in January 2017 showing that no water was present in the region at all (Fig. 11).

The first reporting of the application of chemical molluscicides to open freshwater bodies in Zanzibar was by Macdonald et al. (1968b). It was detailed that between May 1966 and June 1967, all transmission sites in the high-transmission areas around Donge School, Unguja, were systematically

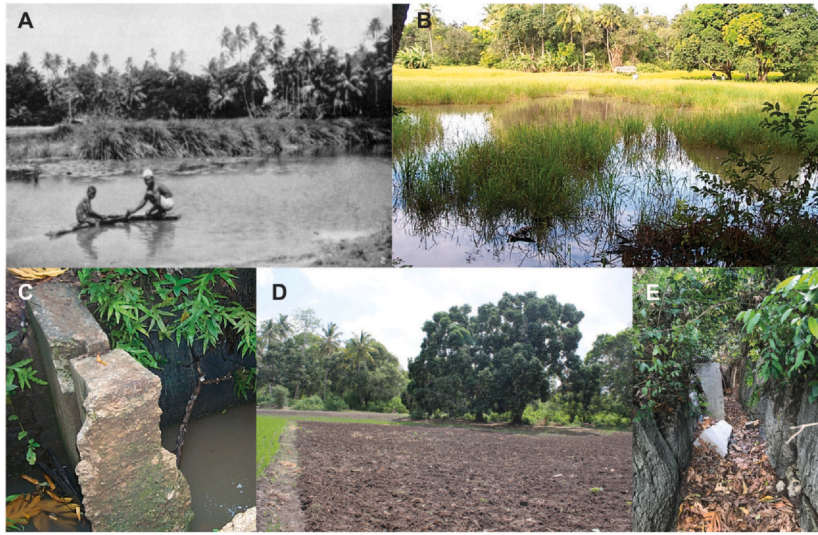


Fig. 11 Photographs from Muyuni pond in southern Unguja, before the construction of 'Mozley's ditch' in 1937/1938 (A) (Mozley, 1939), in May 2001 demonstrating the change in vegetation due to the seasonality of the pond due to the ditch presence (B, C), and again in January 2017 when the pond (D) and ditch (E) are dry and the area can be ploughed as arable land (B, C: personal collection, Russell Stothard; D, E: personal collection, David Rollinson).

inspected for snails, and if snails were present, the molluscicide niclosamide (Bayluscide) was applied.

Almost a decade later, in a mission report by the WHO from a visit to Zanzibar in 1975, Mccullough and Krafft (1976) recommended that in areas with ≥20% *S. haematobium* infection prevalence among 6–16 year old boys, snail control should be implemented alongside preventive chemotherapy and health education and that this should be conducted in close collaboration with local authorities (see sections Treatment interventions, and Behaviour change intervention). Moreover, conducting malacological surveys before and after the use of the molluscicides niclosamide (Bayluscide) or *N*-tritylmorpholine (Frescon) was recommended to assess the presence and number of intermediate host snails around the snail control intervention. The suggestion of this approach was reiterated two years later by Mccullough (1978) and by 1979, a concrete plan was outlined for molluscicide to be introduced as part of a WHO feasibility study for schistosomiasis control methods in Zanzibar (Mccullough et al., 1979). It was suggested that niclosamide should be sprayed in water bodies that are

frequently used by the local population and appear favourable for snail host populations. The first application of molluscicides within the feasibility study was scheduled for the dry season of 1982, and the shehia Kinyasini in the north of Unguja was chosen as the project area due to the high *S. haematobium* prevalence recorded in the general community (46.5%) in 1981 (Mott et al., 1983, Mgeni, 1984b). In 1983, niclosamide (Bayluscide) was applied twice to the seven main *S. haematobium* transmission sites in Kinyasini (Mott et al., 1983, Mgeni et al., 1990), and although observations indicated that an effective snail killing had been achieved (Mott et al., 1983), no quantitative results were recorded on the impact of the intervention on the snail presence/abundance or how this impacted human infection (Mgeni et al., 1990).

While there are no more reports about snail control on Unguja in the 20th century, snail control using niclosamide was conducted on Pemba in December 1994 and February 1995 in all ponds and streams in areas with a *S. haematobium* prevalence >20% (Albonico, 1995). Unfortunately, no information was published about the effectiveness of the intervention and whether the intervention covered the whole island or only specific study areas.

11.2 Large-scale snail control interventions

Niclosamide, the widely used molluscicide (World Health Organization, 2019), was first applied in Zanzibar for snail control purposes on a large scale and over a period of multiple years during the ZEST study from 2012 to 2017 (Knopp et al., 2012, King et al., 2015). In a cluster-randomised trial, the effect of snail control plus biannual MDA was assessed in 30 shehias, 15 shehias per island (Knopp et al., 2012, Knopp et al., 2015b). In each shehia, all known human water contact sites (HWCSs), a total of 121 on Unguja and 167 on Pemba, respectively, were surveyed for snails multiple times per year (Allan et al., 2020). At each HWCS visit, two trained field workers searched for 15 minutes for *Bulinus africanus* species group snails, and if present, niclosamide was applied to reduce the amount of potentially infected intermediate host snails in the treated freshwater bodies (Fig. 12).

Over the five years of implementation from 2012 to 2016 in a total of 30 shehias on both islands, the financial costs for snail control measures were estimated at US$ 129,377 (Salari et al., 2020). These costs were mainly driven by personnel costs and excluded the costs for donated niclosamide. By the end of the trial, the overall *S. haematobium* prevalence on the islands was successfully lowered to below 2% in adults and in

Fig. 12 Malacological survey during the SchistoBreak project in Pemba in 2021 (A), preparing niclosamide as molluscicide during the SchistoBreak project in Pemba in 2021 (B) and during the ZEST project in 2014 in Unguja (C), application of niclosamide with a petrol pump sprayer (D) and with a backpack sprayer during the SchistoBreak project in Pemba in 2021 (E) (A, B, D, E: personal collection, Lydia Trippler; C: personal collection, Tom Pennance).

schoolchildren, respectively. However, due to this low prevalence in the final study year, the data was not statistically powered to detect small but potentially important biological effects that snail control may have had on reducing *S. haematobium* prevalence (Knopp et al., 2019b, Knopp et al., 2019c).

After the ZEST trial had been completed in 2017, snail control was implemented in two shehias on Pemba during the China Aid Schistosomiasis Control Project, running from 2017 to 2019. The associated study aimed to evaluate the adverse effects and efficacy of Chinese formulations of niclosamide (Yang et al., 2019, Zhou et al., 2019, Xing et al., 2021). For this purpose, four water bodies within the study area were surveyed for 15 minutes for the presence and number of *B. globosus* (He et al., 2019, Xing et al., 2021). Following this, the Chinese-made niclosamide was applied up to a 15 meters area off the edge of each water body with a single cylinder engine and a closed impeller pump (Xing et al., 2021). Snail surveys assessing the presence and number of *B. globosus* were

repeated one month and three months after the snail control intervention. The application of Chinese molluscicides was repeated in case of persistent *B. globosus* abundance (Xing et al., 2021). The results showed a decrease in the number of snails in the four water bodies, from 1,043 snails before molluscicide to 143 snails three months after the first application (Zhou et al., 2019, Xing et al., 2021).

Currently, as part of the SchistoBreak study mentioned previously (2020–2024), snail control with niclosamide is being trialled as part of a multi-intervention package in areas with a high transmission of *S. haematobium* (Trippler et al., 2021b). Furthermore, snail control is implemented as part of a reactive surveillance-response interventions in areas with a low transmission of *S. haematobium*. In high transmission shehias, two trained field-workers search for snails in all known HWCSs for ten minutes four times a year. In low transmission shehias, two field-workers search for snail species for ten minutes in all HWCSs, which had been frequented by participants of the study that tested positive for *S. haematobium* and/or microhaematuria. At each water body with a confirmed presence of the intermediate host snail *B. globosus*, niclosamide is applied to the shoreline with a plastic backpack sprayer or a petrol pump machine (Fig. 12), depending on the size and environment of the water body. While results on the potential impact of niclosamide on snail populations and human infections are not yet available, there have been some interesting findings at baseline. In line with studies conducted on Unguja in 2007 and 2014, the SchistoBreak study showed that proximity of schools and households to open freshwater bodies, that are habitat to *B. globosus,* is a major risk factor for *S. haematobium* infection in students and residents in northern Pemba (Rudge et al., 2008, Pennance et al., 2016, Trippler et al., 2022b).

Unlike in Mozley's times, the SchistoBreak study showed that the population in Zanzibar is nowadays not only accepting snail control with niclosamide, but even asking for 'applying medicine to the water' to reduce snail populations or simply to 'kill the snails' (Trippler et al., 2022b). This finding is also an indication that some people in the local communities are aware of how schistosomiasis is transmitted. However, while people ask for snail control and the WHO recommends the intervention with niclosamide for all endemic areas, one must keep in mind that snail control with niclosamide bears certain challenges. These challenges not only include the need of heavy equipment and material, which may have to be carried to often remotely located open freshwater bodies (Campbell et al., 2020), but also the toxicity of the niclosamide, not only for snails, but also for other

freshwater invertebrates, amphibia and fish (World Health Organization, 2022). Hence, there is a possibility that it could interfere with and disrupt the wider aquatic ecosystem.

11.3 Outlook

Challenges of snail control as described above, including the toxicity of niclosamide, underline the need to find alternative solutions. Substitutes may include environmentally friendly and organic substances that still have a molluscicidal effect, such as the plant extracts that Mozley conducted early experiments with and that continue to be an area of active research, although with no real candidates yet established (Mozley, 1939, Pereira et al., 2020). Although there is little evidence for chemical resistance to molluscicides such as niclosamide (Dai et al., 2015), there is a potential risk that resistance, or at least increased tolerance may develop with repeated use (Sullivan et al., 1984). New compounds or active ingredients that may be effective, and ideally more specific to snails, are necessary (World Health Organization, 2019). To ensure the targeted use of niclosamide, it is crucial to align its application with malacological surveys and only use chemical snail control in freshwater bodies where the presence of intermediate host snails has been confirmed. This approach has been followed consistently over the past 100 years in Zanzibar. However, there is the possibility that snails can be overlooked during malacological surveys. Hence, new methodologies such as testing the water for environmental DNA (eDNA), or schistosome larval stages that would simultaneously be captured by water filtration and downstream PCR/qPCR or eDNA metabarcoding approaches, could assist in identifying water bodies in Zanzibar where transmission is taking place. Such an approach has been tested in a study in Kenya where *S. mansoni* eDNA presence in freshwater bodies was successfully identified using a qPCR assay (Sengupta et al., 2019).

Alternatives to plant extracts or niclosamide may be the introduction of natural predators of snails such as freshwater crabs, crayfish, fish and ducks (Mansfield-Aders, 1928, Mkoji et al., 1999, Sokolow et al., 2015). Gene drive systems, such as Clustered Regularly Interspaced Short Palindromic Repeats, known as CRISPR, which can be recruited to favour the selection of genes in vectors to confer resistance to particular pathogens are still very much in their infancy, although have been demonstrated in other vector-borne diseases (Hammond et al., 2016). With new candidate snail immune genes continually being identified (Tennessen et al., 2020), and a growing understanding that susceptibility of intermediate host snails can

change based on schistosome strains (Spaan et al., 2023), it is certainly many years until the open field release of genetically modified snails may occur (Famakinde, 2018, Maier et al., 2019).

What has been clearly demonstrated from the several studies from Zanzibar, and elsewhere, is that MDA alone is not sufficient to reach interruption of schistosomiasis transmission (Ross et al., 2014, Karanja et al., 2017, Knopp et al., 2019c). Hence, unless infrastructural improvements provide alternative options for using natural open freshwater bodies for agriculture, daily chores, bathing and swimming, or until major socio-economic developments occur, snail control, as part of a multidisciplinary intervention approach, remains essential for achieving interruption of *Schistosoma* transmission in Zanzibar (Knopp et al., 2019c, Trippler et al., 2021a, Kura et al., 2022).

12. Behaviour change interventions

12.1 From health education to behaviour change interventions

Human behaviour plays a central role in the transmission and prevention of schistosomiasis. Interventions that promote and enable behaviour change in populations at-risk, and aim to reduce water contact, improve sanitation and increase treatment compliance, are an important complement to praziquantel treatment and snail control (World Health Organization, 2022). Behaviour change interventions should be designed as knowledge and information dissemination by health education, coupled with '*carefully designed interventions that lead to the ability & motivation to change behaviour*' (Global Schistosomiasis Alliance, see www.eliminateschisto.org, access date: 19.01.2023). Moreover, behaviour change interventions should be tailored to the local setting of the at-risk population acknowledging the cultural, demographic and epidemiological context.

In Zanzibar, health education for urogenital schistosomiasis control was first mentioned by Mansfield-Aders (1928) in the Zanzibar Protectorate report from 1927, where he suggested the creation of short pamphlets explaining the disease and its aetiology, and the danger of bathing and washing clothes in swamps. Although it is not reported whether the pamphlets were ever created, the erection of signs near certain water bodies warning of the danger of bathing and the risk of acquiring schistosomiasis was reported three years later (Zanzibar Protectorate, 1930–1934).

Several decades later, health education, integrated with preventive chemotherapy and snail control, was suggested in a WHO mission report from 1976 in preparation of a schistosomiasis control programme on the Zanzibar islands (Mccullough and Krafft, 1976) (see sections Treatment interventions, and Snail control interventions). The use of local broadcasting services and television was suggested to cover a great area and reach many individuals. In 1978, within another WHO mission report, behaviour change interventions were promoted as a revolutionary tool for schistosomiasis control in Zanzibar (Mccullough, 1978). It was stressed that all interventions should be highly authentic and relatable for the population, such as locally produced films or informal talks in the villages rather than sophisticated knowledge dissemination (Mccullough, 1978). These insightful recommendations meant moving away from solely a top-down health education approach, towards promoting the engagement and collaboration with the local population. Subsequently, in 1979, it was proposed to seek advice from embassies and international agencies regarding health education activities as well as to include local groups in their implementation, such as the Youth League or women's groups (Mccullough et al., 1979). One publication by Mgeni et al. (1990) covered a preventive chemotherapy study implemented between 1981 and 1983 that documented health education sessions and improvement of safe water supplies in Kinyasini school, Unguja, although without indicating their potential impact. A study by Savioli et al. (1989a) documented that in 1986, as part of a programme to control urinary schistosomiasis on Pemba, schoolteachers were trained in collaboration with the Zanzibar Ministry of Education, emphasising the human part of the transmission cycle and the responsibility of people to interrupt the transmission cycle. Besides the publication by Mgeni (1990) and Savioli et al. (1989a), no sources are available elaborating whether the behaviour change measures recommended in Mccullough and Krafft (1976), Mccullough (1978) and Mccullough et al. (1979), were implemented in Zanzibar in the following years. In 1994, in a WHO mission travel report Savioli (1994) documented that the Zanzibar MoH had started a school health education campaign in the northern district of Unguja. Although schistosomiasis was not specifically mentioned in the report, the campaign aimed to cover the impact of sanitation on the health of schoolchildren (Savioli, 1994). However, no information about the starting year, the precise location and the outcome of the health education campaign are available.

In 2006, a pilot research study started in five primary schools on Unguja assessing the impact of a cartoon booklet called 'Juma na Kichocho' (Eng.: 'Juma and schistosomiasis') on the schistosomiasis-related knowledge and

attitude of schoolchildren (Stothard et al., 2006b). The booklet, which was designed in collaboration with a local artist and put into production by the support of the WHO, contained the story of a young boy learning about the symptoms and signs of schistosomiasis, the transmission of *S. haematobium*, and about preventive measures for not acquiring and spreading the infection. After introducing the booklet to the children, the teachers were guiding the children through the booklet for 10–15 minutes per day for one week. Before the onset of teaching and after the one week of implementation, childrens' knowledge and attitudes were assessed, and a significant increase in correct responses was observed. After one additional year in which teachers of participating schools were requested to continue using the booklet within their curricula, the children were surveyed again with the same questionnaire (Stothard et al., 2016). Although the average knowledge of all participants did not change significantly after one year of using the booklet, a significant increase of knowledge about schistosomiasis was observed in some schools. The WHO has since revised and adapted 'Juma na Kichocho' into six different languages, made possible through a financial grant from Merck-KGaA (Germany), and is now freely available in a modernised educational format entitled 'Bambo has Bilharzia: What Children Should Know about Bilharzia' (www.who.int/publications/i/item/9789241501903).

12.2 Large-scale behaviour change interventions

Behaviour change measures were applied for the first time at a large scale in 15 schools and 15 communities on Unguja and Pemba, respectively, during the ZEST cluster-randomised trial from 2012 to 2017 (Knopp et al., 2012, Knopp et al., 2015b). The measures were created following a human-centred design approach, which is based on the idea that community members know best which solutions are most suitable and implementable to overcome their own setting-specific difficulties, and consists of three major phases: 'hear', 'create', and 'deliver' (Person et al., 2016b). Following the human-centred design approach starting with 'hear', schistosomiasis-related focus group discussions and in-depth interviews were conducted with children, community leaders, teachers and parents in seven communities in Zanzibar in 2011 to inform the behavioural interventions for ZEST (Person et al., 2012, Knopp et al., 2013a, Person et al., 2016a, Person et al., 2016b). The results revealed poor schistosomiasis-related knowledge and practices in the participants, such as perceiving schistosomiasis as a disease of women only, or frogs being part of the schistosomiasis

transmission life cycle, or not seeking treatment other than plant-based teas due to the cost connected to medical treatment (Person et al., 2012, Knopp et al., 2013a, Person et al., 2016a, Person et al., 2016b).

Based on findings from 'hearing', researchers and community participants co-designed the behaviour change interventions tailored to the local setting in a 'creative' workshop. By the end of the workshop, participants came up with the following suggestions, which formed the basis for 'delivering' the behaviour change interventions in the ZEST study: (i) class-room based interactive education, (ii) school-based Kichocho Safe Play Day events, (iii) male and female urinals, and (iv) washing platforms (Person et al., 2016b).

Within the 'deliver' phase, implementation of these interventions followed a stepwise approach. First, trainings were conducted with school-teachers and religious teachers to increase their knowledge about schistosomiasis and to provide them with new ideas and tools on how to teach schoolchildren and other teachers about preventive behaviour (Person et al., 2016b). One year after the initial training, the teachers had trained another 678 of 761 registered teachers in methods on how to include preventive behaviour on schistosomiasis infection in their curriculum and 27,819 children had received educational training about schistosomiasis (Person et al., 2016b). By the end of the ZEST project, 291 'Kichocho Days' reaching more than 150,000 children, several among them over multiple years, were conducted in primary and Islamic schools (Knopp et al., 2019c). These public outreach days focussed on conveying health education messages via playing and watching safe games that all contained questions and answers about schistosomiasis (Celone et al., 2016, Person et al., 2016b, Knopp et al., 2019c). Engaging Islamic schools and their teachers in the behavioural activities was deemed particularly crucial due to the significant influence the Islamic teachers have within the societies of the islands and serve therefore as key individuals in the change making process (Celone et al., 2016). To disrupt the *S. haematobium* life cycle at the stage where the eggs enter freshwater, 58 male and female urinals were built close to open freshwater bodies in a total of 29 communities (Person et al., 2016b, Knopp et al., 2019c). The urinals were painted with health-seeking instructions for individuals urinating blood, and community members were educated in meetings about the importance of using the urinals instead of urinating into the open freshwater bodies with regard to the schistosomiasis life cycle (Person et al., 2016b). Last, to target individuals using open freshwater bodies to wash their clothes and/or dishes and hence put

themselves at risk for a *S. haematobium* infection, an alternative was provided by installing washing platforms in close proximity or directly connected to safe water sources in the 30 study communities (Fig. 13) (Person et al., 2016b, Knopp et al., 2019b).

The washing platforms created a safe space to wash and socialise, and both men and women used them at different times of the day (Person et al., 2016b). In total, 46 washing platforms were constructed in the 30 study communities (Knopp et al., 2019b). After the five years of implementation of behavioural interventions within the ZEST project, questionnaires were conducted with children of four schools where behaviour change measures were implemented during the project and four schools where the measures were not implemented aiming to compare the schistosomiasis-related knowledge, attitudes and practices of the children (Campbell et al., 2020, Person et al., 2021). The results showed very promising changes of self-reported behaviour in those children who participated in the behaviour change activities during the ZEST project versus those children who did

Fig. 13 Washing platform built during the SchistoBreak project in use on Pemba (personal collection, Lyndsay Taylor). Washing platforms were built neighbouring safe water sources to reduce the schistosome exposure of residents that previously used open freshwater bodies to wash their clothes.

not participate in the intervention: 49.9% versus 5.8% of the children stopped washing laundry/dishes in open freshwater bodies, 49.9% versus 4.2% stopped bathing in stream/ponds and 40.8% versus 10.8% stopped playing in streams/ponds (Person et al., 2021). Similar to the results on snail control, however, behaviour change measures did not significantly boost the effect of biannual MDA for the reduction of *S. haematobium* infections in Zanzibar as determined in the ZEST cluster-randomised trial (Knopp et al., 2019c). The total financial costs for the implementation of the behaviour change measures in 30 shehias on both islands from 2012 to 2017 were estimated at US$ 265,000. As was the case for the financial costs for snail control, the major driver were personnel costs (Salari et al., 2020).

As the behaviour change measures co-designed with the community within the ZEST project were considered as important complementary measures for schistosomiasis elimination when tailored to focal endemicity and applied for a longer period, several of the interventions are being implemented in the ongoing SchistoBreak project from 2020 to 2024 in the north of Pemba (Trippler et al., 2021b). In SchistoBreak, education sessions for teachers and 'Kichocho Days' are conducted once per year in all primary, secondary and Islamic schools that are located in areas considered transmission hotspots (Trippler et al., 2021b). Moreover, in all hotspot communities, two washing platforms have been built in close proximity to safe water sources. The respective communities have also been visited once a year to encourage the use of the washing platforms and to provide general information about *S. haematobium* transmission and prevention measures. Schistosomiasis-related knowledge, attitudes and practices of schoolchildren and adult community members will continue to be assessed within annual cross-sectional school- and household-based surveys. It is hoped that future analyses will reveal the potential impact of the behavioural interventions combined with biannual MDA and snail control on the *S. haematobium* prevalence in hotspot areas, and also assess the sustainability of knowledge, attitudes and practices gained by the behaviour change measures in schools and communities.

With its comprehensive and large-scale behaviour change interventions, the ZEST project was a pioneer in citizen science and transdisciplinary research, where motivating and engaging interventions for schistosomiasis elimination were co-designed together with the local population.

12.3 Outlook

The behaviour change interventions and findings from the operational research conducted on the Zanzibar islands, and those from other regions

in sub-Saharan Africa (Ejike et al., 2017), might have contributed to the addition of guidelines for behaviour change measures being added to the WHO guidelines on control and elimination of human schistosomiasis alongside snail control (World Health Organization, 2022). The guidelines emphasise the importance of behaviour change interventions for the reduction of transmission of *Schistosoma* spp. in endemic areas in the future. Studies in Zanzibar have shown that the compliance with MDA in the adult population is often considerably low (Knopp et al., 2016, Knopp et al., 2019c, Trippler et al., 2022b). Therefore, behaviour change activities should continue among at-risk populations in Zanzibar to improve the awareness of the protective effects that interventions against a *S. haematobium* infection have, and additionally include influential individuals of the society, such as teachers of Islamic schools and religious leaders (Celone et al., 2016). Providing education and the appropriate resources can also help to change people's behaviour to avoid infection and transmission by reducing their contact with potentially contaminated freshwater and to increase treatment-seeking behaviour. However, a change in behaviour can only occur when locally accepted alternatives to open freshwater bodies are accessible. Hence, close collaboration between educational, health, water and environmental sectors is warranted for ultimate success in schistosomiasis control and elimination.

13. Surveillance-response interventions

13.1 Implementation of surveillance-response interventions in Zanzibar

Surveillance is understood as 'the ongoing systematic collection, analysis, and interpretation of outcome specific data for use in planning, implementing and evaluating public health policies and practices' (World Health Organization, 2006). In combination with surveillance, public health response packages can serve as interventions for disease elimination in different endemic settings (Zhou, Bergquist, 2013, Bergquist et al., 2015). While the WHO (2006) has very good guidelines for communicable disease surveillance-response systems, and the importance of a robust surveillance system and of post-transmission surveillance for schistosomiasis control and elimination is mentioned several times in their guidelines on human schistosomiasis from 2022 (World Health Organization, 2022), there is no specific recommendation yet on when, where and how to

conduct pre- and post-elimination surveillance-response for schistosomiasis. However, as an alternative to large-scale MDA without prior diagnosis, in communities with a prevalence <10%, the WHO (2022) now recommends the implementation of test-and-treat as an intervention if there has not been a preventive chemotherapy programme before.

In Zanzibar, there has been plenty of evidence of a pronounced focality of *S. haematobium* transmission, resulting in low- and high-prevalence shehias (Macdonald and Forsyth, 1968, Rudge et al., 2008, Stothard et al., 2009a, Knopp et al., 2019b, Trippler et al., 2021a, Trippler et al., 2022b). After decades of intense control and elimination interventions, there were only a few shehias with high *S. haematobium* prevalence remaining in 2020, whereas the majority of shehias on the Zanzibar islands had a *S. haematobium* prevalence <2% (Trippler et al., 2021b). For these low prevalence shehias, it was felt that large-scale MDA was no longer justifiable, since it would result in an overtreatment of a mostly healthy population, potential treatment fatigue, and an inadequate use of praziquantel and other resources (Trippler et al., 2021b). Hence, it was suggested to tailor interventions to the local micro-epidemiology and to introduce new methods for surveillance and response in the low prevalence shehias to maintain the gains made and to advance towards interruption of transmission.

To gather scientific evidence for the suitability of surveillance-response as an intervention for schistosomiasis elimination, the ongoing SchistoBreak study is implemented in the north of Pemba from 2020 to 2024 (Trippler et al., 2021b). While biannual MDA, snail control and behaviour change interventions are implemented in high transmission shehias (see sections Treatment interventions, Snail control interventions and Behaviour change interventions), the surveillance-response approach is investigated in areas with a *S. haematobium* prevalence <2.0% in community-based surveys and <3.0% in school-based surveys (Trippler et al., 2021b). The project's primary aim is 'to quantify the sensitivity of an adaptive surveillance-response approach for its ability to detect *S. haematobium* infected individuals in low prevalence areas to trigger an appropriate intervention response' (Trippler et al., 2021b). The surveillance-response interventions include active, reactive and passive surveillance implemented in schools, households, water bodies and health facilities, respectively, coupled with responses consisting of treatment of *S. haematobium*-positive individuals and snail control, as described below.

During active surveillance, all children in grades 3–5 in the biggest public primary school and in one Islamic school in each low prevalence

shehia in the study area are tested at least once a year for *S. haematobium* infection by urine filtration and for microhaematuria by reagent strips. Every child that tests positive is treated with a single oral dose of praziquantel (40 mg/kg body weight) with the aid of a dose pole.

For reactive case detection, each child who tested positive is tracked to its home by a member of the research team, and all household members present at the visit are invited to provide a fresh urine sample to be tested for microhaematuria at the point of care and offered a single dose of praziquantel if the sample is positive. In addition, all children who initially tested positive are tracked to the open freshwater bodies they use regularly. At the water bodies, all individuals present at the time of the visit are offered testing with reagent strips for microhaematuria at the point of care and, if tested positive, offered treatment with praziquantel. Surveys for the intermediate host snail *B. globosus* are also carried out in the same water bodies, and, if the surveys confirm the presence of *B. globosus*, snail control with niclosamide is conducted (see section Snail control interventions).

For passive surveillance, staff of primary health care units located in the study area are trained on the prevention and transmission of schistosomiasis, the diagnosis of microhaematuria with reagent strips and the treatment with praziquantel by staff of the research team. The primary health care units are provided with Hemastix reagent strips to test all patients for microhaematuria who report symptoms of urogenital schistosomiasis. Moreover, the primary health care units are equipped with praziquantel to treat the haematuria-positive patients. At least once a month, the primary health care units report the number and details of patients who presented with symptoms, if they were tested for microhaematuria and if they were treated with praziquantel, to the study team.

13.2 Outlook

Currently, the SchistoBreak project is still ongoing, but it is expected that informative results regarding test–treat–track and surveillance-response will be published in the coming years. The results will help fill some of the existing evidence gap on surveillance-response methodologies for schistosomiasis elimination. The results should also indicate whether: (a) risk-based surveillance combined with a targeted test–treat–track approach can identify *S. haematobium*-positive individuals in Zanzibar, (b) if the surveillance approach can suppress recrudescence of transmission and infection in low prevalence areas where MDA has been stopped, and (c) if the targeted study shehias continue to move towards interruption of *S. haematobium* transmission.

14. Conclusion

Whilst the main Zanzibar islands have undergone many political and cultural developments over the past 100 years, they have also become 'model islands' for research into several NTDs, including schistosomiasis, which has been demonstrated throughout the findings of this review. Over the last decade, the overarching aim for public health targets has moved from controlling schistosomiasis morbidity to the complete elimination of *S. haematobium* from the islands. With a low overall prevalence of urogenital schistosomiasis, and the availability of chemotherapeutics, diagnostics and interventions targeted to break the life cycle of the parasite pioneered on the islands, elimination has become an achievable goal. The review highlights the spatial heterogeneity that has been observed on the islands since the 1960s. This heterogeneity is evident through many low prevalence and low intensity areas co-existing with a few hotspot areas on Unguja and Pemba. Despite an overall heavy-intensity infection prevalence of <1%, these hotspot areas showed a heavy-intensity infection prevalence >1%, thus impeding the attainment of the WHO goal 'elimination as a public health problem', for several years. Therefore, the revision in the WHO's NTD roadmap definition of 'elimination as a public health problem' in 2020 including the removal of 'in all sentinel sites' from the goal, was a positive and commendable change for the Zanzibar islands and provided them with the opportunity to be officially declared an elimination setting, now aiming for 'interruption of *S. haematobium* transmission'. In reviewing and summarising the vast extent of knowledge generated with research and interventions in Zanzibar over the past century in this systematic review, we highlight the key points that need to be addressed to reach interruption of *S. haematobium* transmission and provide below future avenues for research that will help achieve the goal.

While the role of *B. globosus* as an intermediate host snail for urogenital schistosomiasis in Zanzibar has been proven many times, there remain many open research questions. First and foremost, the role of endemic *B. nasutus* as a second viable intermediate host snail species needs to be further clarified with a thorough experimental investigation of compatibility with *S. haematobium* populations. Second, detailed mapping, molecular identification and xenomonitoring to assess exposure and compatibility of all endemic *Bulinus* species (including *B. globosus* and *B. nasutus* and the three species of the *B. forskalii* species group) to schistosomes is essential. Considering the recent establishment of the cattle

parasite *S. bovis* in some regions, the associated risk of *S. haematobium–bovis* hybrid introduction/emergence and the expanded intermediate host range that can occur because of this also needs to be closely monitored. Due to daily travel between mainland Africa, specifically coastal Tanzania and Kenya, and the Zanzibar islands, the concern regarding introduction also pertains to *S. haematobium* strains that are compatible with *B. nasutus* on Zanzibar. The identification of a single infected snail carrying a mainland group *S. haematobium* parasite is hinting at the potential introduction of new strains that may contribute towards expanding the intermediate host range of *S. haematobium*. In future elimination efforts, it may become necessary to screen incoming individuals for infection, to limit chances of reintroduction of *S. haematobium*, or bring strains that will further complicate transmission.

Besides molecular analyses of (potential) intermediate host snails, molecular analyses of the *S. haematobium* populations may be valuable in monitoring any genetic changes of the parasite, such as selection or advantageous adaptions for survival. Whole genome-based analyses can further identify genotypic and phenotypic differences, which will provide insights into how schistosomes might change under high selection pressures. Regular sampling of parasite populations for the purposes of future molecular analyses is paramount to track signatures of selection, development of drug resistance/tolerance, importation of *S. haematobium* from other endemic areas, compatibility with several intermediate snail host species and interspecies hybridisation, all of which may complicate reaching and maintaining elimination. Given the constant risk of first generation hybridisation between *S. haematobium* and *S. bovis* on the islands, or the introduction of new strains from elsewhere, it is crucial to continue collecting and conducting genotypic analysis of the schistosomes, since changes in the parasite genetics could potentially alter the disease epidemiology on the islands (Emery et al., 2012).

As schistosomiasis prevalence has varied over time and space, it has provided a platform to evaluate the type of diagnostics that are appropriate in different stages of schistosomiasis control through to elimination levels. Basic diagnostics, such as detecting haematuria have proven to be economically and logistically efficient in guiding early interventions when prevalence and intensity of infections are highest. Urine filtration followed by egg microscopy provides a useful follow-up measure during preventative chemotherapy campaigns to control schistosomiasis, as it allows for both the prevalence and intensity of infection to be recorded easily and to assess the impact of treatments when more subtle differences (such as

reduced infection intensity) are important. However, fundamental issues exist in the use of currently available diagnostics in elimination settings where low prevalence and light intensity infections are the majority and cannot be consistently detected. Therefore, sensitive and specific diagnostic tools, ideally rapid diagnostic tests for the use at the point of care, are urgently needed for early case detection to prevent resurgence in low transmission areas and to respond immediately to outbreaks. Sensitive and specific diagnostics are also essential for assessing the residual acute and chronic morbidity associated with schistosome infections of individuals residing in formerly high endemic settings that are now considered to have achieved elimination as a public health problem according to the WHO definitions. The extent of morbidity in lightly infected individuals, and urogenital tract pathologies including FGS and MGS, that may be associated with a present or past *S. haematobium* infection urgently needs detailed assessment.

In terms of interventions, Zanzibar has been at the forefront of implementing systematic, large-scale treatment programmes since the 1980s. Repeated and constantly refined MDA schemes have led to the elimination of urogenital schistosomiasis as a public health problem in most areas in Zanzibar, although transmission has not yet been interrupted. While many shehias on the Zanzibar islands show a low transmission of *S. haematobium*, there are other shehias with persistent moderate to high transmission. To reach interruption of *S. haematobium* transmission in these remaining hotspots, most likely a multidisciplinary intervention approach consisting of treatment, snail control and behaviour change interventions coupled with improvements in the sanitary infrastructure is needed. Whether targeted surveillance-response interventions in low transmission shehias can avoid overtreatment of a mostly healthy population and treatment fatigue, and at the same time prevent resurgence of transmission and infection will be shown by ongoing and future studies in Zanzibar. The current SchistoBreak study, being conducted in the north of Pemba, will contribute evidence for the suitability of surveillance-response and test–treat–track–test–treat interventions for schistosomiasis elimination in low transmission shehias in Zanzibar and elsewhere, informing NTD programme managers and providing evidence for future WHO recommendations.

In addition to treatment, snail control has been proven to be an important part of a multidisciplinary intervention approach and has a critical role to play in ultimately interrupting *S. haematobium* transmission in

Zanzibar and beyond. However, there is a need to find environmentally friendly alternatives to currently used molluscicides that can more specifically target the intermediate host snails of *S. haematobium* without disrupting aquatic ecosystems. It is also crucial that behaviour change interventions continue reminding residents of the schistosome-human-schistosome transmission cycle, including how not to contribute towards parasite transmission, the risks associated with infection and the importance of treatment.

Disseminating public health information relating to *Schistosoma* transmission needs to be combined with improved access to safe water sources, which will minimise exposure to potentially contaminated water bodies and also demonstrate long term commitments that promote treatment-seeking behaviour. Infrastructural freshwater developments should include culturally acceptable sanitation and hygiene facilities, but also agricultural schemes to benefit sustainability and food security in Zanzibar. Such agricultural developments have commenced in both Unguja and Pemba with the construction of several dams to create water reservoirs that will provide a consistent means of irrigation, extending the amount of arable land and increasing food production that is necessary to sustain the growing population (Office of Chief Government Statistician Zanzibar and Ministry of Finance and Planning Zanzibar, 2020). However, the emerging water reservoirs, canals and irrigated fields provide new habitats for the intermediate host snails (Fig. 8), and potentially other endemic/invasive freshwater biota that should be closely monitored. These new snail habitats combined with human and cattle activities around these attractive water bodies present ideal conditions for transmission, and will hence be detrimental to Zanzibar's commitment to eliminate urogenital schistosomiasis. On the other hand, with the appropriate infrastructure planning, engagement from stakeholders and multisectoral collaboration, the dams could offer the opportunity to provide a reliable source for safe (aged) piped water to neighbouring communities and increase food supplies.

The Zanzibar MoH together with various stakeholders from different research institutions and non-governmental organisations from different countries have shaped the course of schistosomiasis epidemiology on the Zanzibar islands over the past 100 years, enabling the islands to reach elimination of urogenital schistosomiasis as a public health problem in 2017. Over the years, Unguja and Pemba have served as a test bed for innovative approaches in schistosomiasis control, surveillance and elimination, and the knowledge gained from the related research on the islands has contributed to and shaped the development of recent WHO guidelines

for control and elimination of schistosomiasis. However, the greatest achievement of 100 years of schistosomiasis research and implementation of interventions on Unguja and Pemba is undoubtedly the improved health and well-being of both children and adults on the islands, together with the important lessons provided to the global schistosomiasis community in how to accomplish this. Therefore, the fight against schistosomiasis on Unguja and Pemba should not stop until zero incidence of schistosomiasis is reached and maintained, and while interruption of *S. haematobium* transmission across both islands by 2030 might be an ambitious goal, the laudable efforts of the past shows that much can be achieved by operational research and the practical implementation of integrated interventions.

Acknowledgements

We dedicate this publication to the many people who participated in the numerous studies that have been published about schistosomiasis and related snail research within the last 100 years. Furthermore, we express our gratitude to shehas, village leaders and school and madrassa teachers that have enabled research in their communities and schools. We also want to express our heartfelt appreciation for the unwavering dedication and tireless efforts of staff from the Zanzibar Ministry of Health and the Public Health Laboratory-Ivo de Carneri in the ongoing battle against schistosomiasis. In addition, the review has revealed a multitude of stakeholders committing their time, resources and expertise to combat this devastating disease. We extend our deepest appreciation to these individuals and organisations for their dedication and hard work. We would also like to acknowledge the help from Andy Hardy (Aberystwyth University) for useful discussions on the geological features on Unguja and Pemba that contribute towards describing the distribution of *Bulinus* spp. presented in this review. Finally, we express our thanks to Geoffrey Gobert (Queen's University Belfast) for organising the workshop 'Schistosomiasis in health and agriculture' on 29th May to 1st June 2023 that took place on Unguja, Zanzibar and was funded by the Royal Academy of Engineering, United Kingdom. The workshop provided an excellent platform for the in-depth discussion of this topic, resulting in an improved review.

Disclosure

LT and SK are funded by the Swiss National Science Foundation (SNSF) via the PRIMA grant (PR00P3_179753/1) of Stefanie Knopp. TP is currently funded by National Institutes of Health (NIH) grant (R01AI141862) of Michelle Steinauer, with previous support contributing to this work received from the Natural Environmental Research Council (NERC) GW4 + DTP (NE/L002434/1).

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at https://doi.org/10.1016/bs.apar.2023.06.001.

References

Agnew-Blais, J., Carnevale, J., Gropper, A., Shilika, E., Bail, R., Ngoma, M., 2009. *Schistosomiasis haematobium* prevalence and risk factors in a school-age population of peri-urban Lusaka, Zambia. J. Trop. Pediatr. 56, 247–253.

Alawy, K.S., 1993. Prevalence of Schistosomiasis - Positive Haematuria. *From* WHO Archives B2/370/3TAN.

Alawy, K.S., 1995. Expenses Pemba Island Helminth Control Programme. *From* WHO Archives P2/370/4TAN.

Albonico, M., 1995. Travel Report - Dr Marco Albonico - 5 March 1995 - 15 April 1995. *From* WHO Archives P2_370_3.

Albonico, M., 1997. Travel Report Zanzibar 27 August - 1 October 1997. *From* WHO Archives.

Albonico, M., Chwaya, H.M., Montresor, A., Stolfzfus, R.J., Tielsch, J.M., Alawi, K.S., et al., 1997. Parasitic infections in Pemba Island school children. East. Afr. Med. J. 74, 294–298.

Ali, S.M., Albonico, M., Montresor, A., Molyneux, D., Savioli, L., 2015. Impact of preventive chemotherapy on burden of disease due to helminthiasis: The lesson from Zanzibar. Trop. Med. Int. Health 20, 42–43.

Allan, F., Ame, S.M., Tian-Bi, Y.N.T., Hofkin, B.V., Webster, B.L., Diakité, N.R., et al., 2020. Snail-related contributions from the schistosomiasis consortium for operational research and evaluation program including xenomonitoring, focal mollusciciding, biological control, and modeling. Am. J. Trop. Med. Hyg. 103, 66–79.

Allan, F., Dunn, A.M., Emery, A.M., Stothard, J.R., Johnston, D.A., Kane, R.A., et al., 2013. Use of sentinel snails for the detection of *Schistosoma haematobium* transmission on Zanzibar and observations on transmission patterns. Acta Trop. 128, 234–240.

Allan, F., Rollinson, D., Smith, J.E., Dunn, A.M., 2009. Host choice and penetration by *Schistosoma haematobium* miracidia. J. Helminthol. 83, 33–38.

Ame, S.M. 2018. Implementation and Evaluation of Strategies for Control of Schistosomiasis and Soil Transmitted Helminthiasis in Pemba Island, Zanzibar.

Anastasiou, E., Lorentz, K.O., Stein, G.J., Mitchell, P.D., 2014. Prehistoric schistosomiasis parasite found in the Middle East. Lancet Infect. Dis. 14, 553–554.

Archer, J., Barksby, R., Pennance, T., Rostron, P., Bakar, F., Knopp, S., et al., 2020. Analytical and clinical assessment of a portable, isothermal recombinase polymerase amplification (RPA) assay for the molecular diagnosis of urogenital schistosomiasis. Molecules 25.

Aregawi, M.W., Ali, A.S., Al-Mafazy, A.-W., Molteni, F., Katikiti, S., Warsame, M., et al., 2011. Reductions in malaria and anaemia case and death burden at hospitals following scale-up of malaria control in Zanzibar, 1999-2008. Malar. J. 10, 46.

Atalabi, T.E., Lawal, U., Ipinlaye, S.J., 2016. Prevalence and intensity of genito-urinary schistosomiasis and associated risk factors among junior high school students in two local government areas around Zobe Dam in Katsina State, Nigeria. Parasit. Vectors 9, 388.

Attili, V.R., Hira, S.K., Dube, M.K., 1983. Schistosomal genital granulomas: a report of 10 cases. Br. J. Vener. Dis. 59, 269–272.

Austin, T.A., 1955. A survey of conditions in the Zanzibar Protectorate. *From* WHO Archives WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 135–143.

Barakamfitiye, D., 1991. Integrated control of tropical diseases in Pemba, Zanzibar, Tanzania. *From* WHO Archives WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 531–532.

Bergquist, R., Yang, G.-J., Knopp, S., Utzinger, J., Tanner, M., 2015. Surveillance and response: Tools and approaches for the elimination stage of neglected tropical diseases. Acta Trop. 141, 229–234.

Bevilacqua, N., Pane, S., Vairo, F., Nicastri, E., Paglia, M.G., Ame, S.M., et al., 2012. Accuracy of indirect haemagglutination and western blot assays for the detection of anti-*Schistosoma* antibodies in non-severe febrile patients in two Tanzanian hospitals. Scand. J. Infect. Dis. 44, 453–458.

Bhattarai, A., Ali, A.S., Kachur, S.P., Mårtensson, A., Abbas, A.K., Khatib, R., et al., 2007. Impact of Artemisinin-based combination therapy and insecticide-treated nets on Malaria burden in Zanzibar. PLOS Med. 4, e309.

Boissier, J., Grech-Angelini, S., Webster, B.L., Allienne, J.-F., Huyse, T., Mas-Coma, S., et al., 2016. Outbreak of urogenital schistosomiasis in Corsica (France): an epidemiological case study. Lancet Infect. Dis. 16, 971–979.

Bourguignat, J.R., 1879. *Description de diverses espèces terrestres et fluviatiles et de différents genres de mollusques de l'Égypte, de l'Abyssinie, de Zanzibar, du Sénégal et du centre de l'Afrique,* Paris. J. Tremblay.

Brown, D.S., 1994. Freshwater Snails of Africa and Their Medical Importance, 1st ed. CRC Press.

Brown, D.S., 1995. Systematic malacology: the foundation of snail-schistosome studies. A Status of Research on Medical Malacology in Relation to Schistosomiasis in Africa. Copenhagen.

Buddie, A.G., Rwomushana, I., Offord, L.C., Kibet, S., Makale, F., Djeddour, D., et al., 2021. First report of the invasive snail *Pomacea canaliculata* in Kenya. CABI Agric. Biosci. 2, 11.

Bustinduy, A.L., Randriansolo, B., Sturt, A.S., Kayuni, S.A., Leustcher, P.D.C., Webster, B.L., et al., 2022. An update on female and male genital schistosomiasis and a call to integrate efforts to escalate diagnosis, treatment and awareness in endemic and non-endemic settings: The time is now. Adv. Parasitol. 115, 1–44.

Campbell, C.H., Binder, S., King, C.H., Knopp, S., Rollinson, D., Person, B., et al., 2020. SCORE operational research on moving toward interruption of schistosomiasis transmission. Am. J. Trop. Med. Hyg. 103, 58–65.

Candau, M.G., 1964. Correspondence about agreement between the World Health Organization and the "Bilharziasis Laboratory" in Copenhagen. *From* WHO Archives B2_181_4. 38–41.

Cawston, F.G., 1924. Distribution of snails serving as intermediate hosts of flukes in South Africa. Parasitology 16, 67–68.

Cawston, F.G., 1927a. Some notes on the effect of desiccation upon molluscan disease-carriers. Ann. Trop. Med. Parasitol. 21, 35–38.

Cawston, F.G., 1927b. The snail host for bilharzia in Zanzibar. J. Trop. Med. Hyg. 209.

Celone, M., Person, B., Ali, S.M., Lyimo, J.H., Mohammed, U.A., Khamis, A.N., et al., 2016. Increasing the reach: Involving local Muslim religious teachers in a behavioral intervention to eliminate urogenital schistosomiasis in Zanzibar. Acta Trop. 163, 142–148.

Chan, M.S., Montresor, A., Savioli, L., Bundy, D.A.P., 1999. Planning chemotherapy based schistosomiasis control: validation of a mathematical model using data on *Schistosoma haematobium* from Pemba, Tanzania. Epidemiol. Infect. 123, 487–497.

Chopra, S.A., 1968. Malaria, sickling incidence and bilharziasis in relation to agricultural changes and population structure and its movement in Zanzibar. East. Afr. Med. J. 45, 309–310.

Cioli, D., Pica-Mattoccia, L., Archer, S., 1995. Antischistosomal drugs: past, present, future? Pharmacol. Ther. 68, 35–85.

Cohen, J.E., 1974. Some potential economic benefits of eliminating mortality attributed to schistosomiasis in Zanzibar. Soc. Sci. Med. 8, 383–398.

Colley, D.G., Bustinduy, A.L., Secor, W.E., King, C.H., 2014. Human schistosomiasis. Lancet 383, 2253–2264.

Confalonieri, V., 2009. The Pemba challenge in Tanzania. Quaderni ACP 16, 64–65.

Corstjens, P.L.A.M., Dood, D.E., Knopp, C.J., Clements, S., Ortu, M.N., Umulisa, G., et al., 2020. Circulating anodic antigen (CAA): A highly sensitive diagnostic biomarker to detect active *Schistosoma* infections—improvement and use during SCORE. Am. J. Trop. Med. Hyg. 103, 50–57.

Crowther, A., Veall, M.-A., Boivin, N., Horton, M., Kotarba-Morley, A., Fuller, D.Q., et al., 2015. Use of Zanzibar copal (Hymenaea verrucosa Gaertn.) as incense at Unguja Ukuu, Tanzania in the 7–8th century CE: chemical insights into trade and Indian Ocean interactions. J. Archaeol. Sci. 53, 374–390.

Cunin, P., Tchuem Tchuenté, L.A., Poste, B., Djibrilla, K., Martin, P.M., 2003. Interactions between *Schistosoma haematobium* and *Schistosoma mansoni* in humans in north Cameroon. Trop. Med. Int. Health 8, 1110–1117.

Dai, J.-R., Li, Y.-Z., Wang, W.E.I., Xing, Y.-T., Qu, G.-L., Liang, Y.-S., 2015. Resistance to niclosamide in *Oncomelania hupensis*, the intermediate host of *Schistosoma japonicum*: should we be worried? Parasitology 142, 332–340.

De Carneri, A.C., 1994. Correspondence about building a public health laboratory. *From* WHO Archives P2_370_4ZAN.

De Rosa, E., Person, B., Knopp, S., Muhsin, J., Lyimo, J.H., Kabole, F., et al., 2022. A descriptive qualitative case study of the experiences, perceptions and attitudes of pregnant women on Unguja island, Zanzibar, towards antischistosomal treatment. Acta Trop. 225, 106143.

Deganello, R., 2000. Assessment of the impact of chemotherapy campaigns on the morbidity of urinary schistosomiasis in Pemba Island, Tanzania, Thesis. London School of Hygiene andTropical Medicine.

Doenhoff, M.J., Chiodini, P.L., Hamilton, J.V., 2004. Specific and sensitive diagnosis of schistosome infection: can it be done with antibodies? Trends Parasitol. 20, 35–39.

Dowdle, W.R., 1998. The principles of disease elimination and eradication. Bull. World Health Organ. 76, 22–25.

Ejike, C.U., Oluwole, A.S., Mogaji, H.O., Adeniran, A.A., Alabi, O.M., Ekpo, U.F., 2017. Development and testing of Schisto and Ladders™, an innovative health educational game for control of schistosomiasis in schoolchildren. BMC Res. Notes 10, 236.

Emery, A.M., Allan, F.E., Rabone, M.E., Rollinson, D., 2012. Schistosomiasis collection at NHM (SCAN). Parasit. Vectors 5, 185.

Emery, A.M., Loxton, N.J., Stothard, J.R., Jones, C.S., Spinks, J., Llewellyn-Hughes, J., et al., 2003. Microsatellites in the freshwater snail *Bulinus globosus* (Gastropoda: Planorbidae) from Zanzibar. Mol. Ecol. Notes 3, 108–110.

Evans, D., 1997. UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases (TDR). *From* WHO Archives.

Famakinde, D.O., 2018. Treading the path towards genetic control of snail resistance to schistosome infection. Trop. Med. Infect. Dis. 3.

Fenwick, A., Norris, W., Mccall, B., 2021. A Tale of a Man, a Worm and a Snail. CABI.

Ford, D., Williams, P., 2007. Karst Hydrogeology and Geomorphology. John Wiley & Sons Ltd.

Forsyth, D.M., 1965. Treatment of urinary schistosomiasis - Practice and theory. Lancet 286, 354–357.

Forsyth, D.M., 1966. Treating urinary schistosomiasis in the field: a clinical trial of Ambilhar, a treatment campaign for a school in an endemic area, and a pilot study for a mass-treatment programme. Ann. Trop. Med. Parasitol. 60, 172–181.

Forsyth, D.M., 1969. A longitudinal study of endemic urinary schistosomiasis in a small East African community. Bull. World Health Organ. 40, 771–783.

Forsyth, D.M., 1970. Anaemia in Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 64, 601–606.

Forsyth, D.M., 1971. The intradermal test and the plasma card test: a critical assessment of their reliability for diagnosis *Schistosoma haematobium* infection, and other considerations. Ann. Trop. Med. Parasitol. 65, 505–511.

Forsyth, D.M., Bradley, D.J., Mcmahon, J., 1970. Death attributed to kidney failure in communities with endemic urinary schistosomiasis. Lancet 2, 472–473.
Forsyth, D.M., Macdonald, G., 1966. Urological complications of endemic schistosomiasis in schoolchildren Part 2. Donge school, Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 60, 568–578.
Forsyth, D.M., Rashid, C., 1967a. Treatment of urinary schistosomiasis with Trichlorophone. Lancet 290, 909–912.
Forsyth, D.M., Rashid, C., 1967b. Treatment of urinary schistosomiasis - Practice and theory. Lancet 21, 130–133.
Frandsen, F., Christensen, N.O., 1984. An introductory guide to the identification of cercariae from African freshwater snails with special reference to cercariae of trematode species of medical and veterinary importance. Acta Trop. 41, 181–202.
French, M.D., Rollinson, D., Basáñez, M.G., Mgeni, A.F., Khamis, I.S., Stothard, J.R., 2007. School-based control of urinary schistosomiasis on Zanzibar, Tanzania: Monitoring micro-haematuria with reagent strips as a rapid urological assessment. J. Pediatr. Urol. 3, 364–368.
Gazzinelli-Guimaraes, P.H., Dhanani, N., King, C.H., Campbell, C.H., Aurelio, H.O., Ferro, J., et al., 2018. Accuracy of the WHO praziquantel dose pole for large-scale community treatment of urogenital schistosomiasis in northern Mozambique: Is it time for an update? PLOS Negl. Trop. Dis. 12, e0006957.
Glenn, T.C., Lance, S.L., Mckee, A.M., Webster, B.L., Emery, A.M., Zerlotini, A., et al., 2013. Significant variance in genetic diversity among populations of *Schistosoma haematobium* detected using microsatellite DNA loci from a genome-wide database. Parasit. Vectors 6, 300.
Goatly, K.D., Jordan, P., 1965. Schistosomiasis in Zanzibar and Pemba. East. Afr. Med. J. 42, 1–9.
Gower, C.M., Shrivastava, J., Lamberton, P.H., Rollinson, D., Webster, B.L., Emery, A., et al., 2007. Development and application of an ethically and epidemiologically advantageous assay for the multi-locus microsatellite analysis of *Schistosoma mansoni*. Parasitology 134, 523–536.
Groenendael, L.V., Flinkenflogel, L., Vliet, J.A.V.D., Roovers, E.A., Sterkenburg, S.M.V., Reijnen, M.M.P.J., 2010. Conventional surgery and endovenous laser ablation of recurrent varicose veins of the small saphenous vein: a retrospective clinical comparison and assessment of patient satisfaction. Phlebology 25, 151–157.
Grzych, J.M., Dissous, C., Capron, M., Torres, S., Lambert, P.H., Capron, A., 1987. *Schistosoma mansoni* shares a protective carbohydrate epitope with keyhole limpet hemocyanin. J. Exp. Med. 165, 865–878.
Guidi, A., Andolina, C., Ame, S.M., Albonico, M., Cioli, D., Haji, H.J., 2010. Praziquantel efficacy and long-term appraisal of schistosomiasis control in Pemba Island. Trop. Med. Int. Health 15, 614–618.
Haji, H.J., Mgeni, A.F., 1980. A Study of Prevalence of Schistosomiasis in Pupils of Kinyasini and Mahonda Schools, Northern District, Zanzibar. *From* WHO Archives WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 311–325.
Hammond, A., Galizi, R., Kyrou, K., Simoni, A., Siniscalchi, C., Katsanos, D., et al., 2016. A CRISPR-Cas9 gene drive system targeting female reproduction in the malaria mosquito vector *Anopheles gambiae*. Nat. Biotechnol. 34, 78–83.
Hanelt, B., Brant, S.V., Steinauer, M.L., Maina, G.M., Kinuthia, J.M., Agola, L.E., et al., 2009. *Schistosoma kisumuensis* n. sp. (Digenea: Schistosomatidae) from murid rodents in the Lake Victoria Basin, Kenya and its phylogenetic position within the *S. haematobium* species group. Parasitology 136, 987–1001.
Hardy, A., Mageni, Z., Dongus, S., Killeen, G., Macklin, M.G., Majambare, S., et al., 2015. Mapping hotspots of malaria transmission from pre-existing hydrology, geology

and geomorphology data in the pre-elimination context of Zanzibar, United Republic of Tanzania. Parasit. Vectors 8, 41.
Hatz, C., Savioli, L., Mayombana, C., Dhunputh, J., Kisumku, U.M., Tanner, M., 1990. Measurement of schistosomiasis-related morbidity at community level in areas of different endemicity. Bull. World Health Organ. 68, 777–787.
He, J., Kabole, F., 2021. Schistosomiasis control program in Zanzibar: an overview. In: Yang, K., Mehlhorn, H. (Eds.), Sino-African cooperation for schistosomiasis control in Zanzibar: A blueprint for combating other parasitic diseases. Springer, Cham.
He, M.Z., Li, W., Juma, S., Kabole, F., Xu, D.C., Wang, X.Y., et al., 2019. A Google Earth-based database management for schistosomiasis control in Zanzibar. Geospat. Health 14.
Hoekstra, P.T., Van Dam, G.J., Lieshout, L, V.A.N., 2021. Context-specific procedures for the diagnosis of human schistosomiasis – A mini review. Front. Trop. Dis. 2.
Hoover, C.M., Knopp, S., Hubbard, A., Lewnard, J.A., Leo, D.E., Sokolow, G.A., et al., 2020. Dynamics of parasite aggregation under intense control efforts: insights from the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) study. Am. J. Trop. Med. Hyg. 103, 429.
Iozzi, S., Piva, P., Savioli, L., 1984. Acute intestinal obstruction from a granuloma due to *Schistosoma hematobium* in the appendix. Trop. Geogr. Med. 36, 87–89.
Janovski, N., Douglas, C., 1972. Schistosomiasis (bilharziasis). In: Janovski, N., Douglas, C. (Eds.), Diseases of the Vulva, 1st Amer. ed. Harper & Row, Hagerstown, MD.
Jørgensen, A., Madsen, H., Nalugwa, A., Nyakaana, S., Rollinson, D., Stothard, J.R., et al., 2011. A molecular phylogenetic analysis of *Bulinus* (Gastropoda: Planorbidae) with conserved nuclear genes. Zool. Scr. 40, 126–136.
Kalinda, C., Chimbari, M.J., 2022. Effects of laboratory-induced desiccation on fecundity and survival of *Bulinus globosus* (Gastropoda: Planorbidae). Aquat. Ecol. 56, 143–152.
Kane, R.A., Stothard, J.R., Emery, A.M., Rollinson, D., 2008. Molecular characterization of freshwater snails in the genus *Bulinus*: a role for barcodes? Parasit. Vectors 1, 15.
Karanja, D.M.S., Awino, E.K., Wiegand, R.E., Okoth, E., Abudho, B.O., Mwinzi, P.N.M., et al., 2017. Cluster randomized trial comparing school-based mass drug administration schedules in areas of western Kenya with moderate initial prevalence of *Schistosoma mansoni* infections. PLOS Negl. Trop. Dis. 11, e0006033.
Kayuni, S., Lampiao, F., Makaula, P., Juziwelo, L., Lacourse, E.J., Reinhard-Rupp, J., et al., 2019. A systematic review with epidemiological update of male genital schistosomiasis (MGS): a call for integrated case management across the health system in sub-Saharan Africa. Parasite Epidemiol. Control 4, e00077.
Keller, D., Rothen, J., Dangy, J.P., Saner, C., Daubenberger, C., Allan, F., et al., 2020. Performance of a real-time PCR approach for diagnosing *Schistosoma haematobium* infections of different intensity in urine samples from Zanzibar. Infect. Dis. Poverty 9.
Kent, P.E., Kent, P., Hunt, J.A., Johnstone, D.W., 1971. The Geology and Geophysics of Coastal Tanzania. H. M. Stationery Office.
Kim, J.Y., Sim, S., Chung, E.J., Rim, H.J., Chai, J.Y., Min, D.Y., et al., 2020. Effectiveness of mass drug administration on neglected tropical diseases in schoolchildren in Zanzibar, Tanzania. Korean J. Parasitol. 58, 109–119.
King, C.H., Binder, S., Campbell, C., Colley, D.G., 2015. A 5-year progress report from SCORE, the Schistosomiasis Consortium for Operational Research and Evaluation. Trop. Med. Int. Health 20, 53.
Knopp, S., Ame, S.M., Hattendorf, J., Ali, S.M., Khamis, I.S., Bakar, F., et al. 2019a. Correction to: Urogenital schistosomiasis elimination in Zanzibar: Accuracy of urine filtration and haematuria reagent strips for diagnosing light intensity *Schistosoma haematobium* infections (Parasites & Vectors (2018) 11:(552), Parasites & Vectors, 12, https://doi.org/10.1186/s13071-018-3136-6.

Knopp, S., Ame, S.M., Person, B., Hattendorf, J., Rabone, M., Juma, S., et al., 2019b. A 5-year intervention study on elimination of urogenital schistosomiasis in Zanzibar: parasitological results of annual cross-sectional surveys. PLOS Negl. Trop. Dis. 13, e0007268.

Knopp, S., Corstjens, P.L.A.M., Koukounari, A., Cercamondi, C.I., Ame, S.M., Ali, S.M., et al., 2015a. Sensitivity and specificity of a urine circulating anodic antigen test for the diagnosis of *Schistosoma haematobium* in low endemic settings. PLOS Negl. Trop. Dis. 9, e0003752.

Knopp, S., Mohammed, K.A., Ali, S.M., Khamis, I.S., Ame, S.M., Albonico, M., et al., 2012. Study and implementation of urogenital schistosomiasis elimination in Zanzibar (Unguja and Pemba islands) using an integrated multidisciplinary approach. BMC Public Health 12, 930.

Knopp, S., Mohammed, K.A., Stothard, J.R., Khamis, I.S., Rollinson, D., Marti, H., et al., 2010. Patterns and risk factors of helminthiasis and anemia in a rural and a peri-urban community in Zanzibar, in the context of helminth control programs. PLOS Negl. Trop. Dis. 4, e681.

Knopp, S., Person, B., Ame, S.M., Ali, S.M., Hattendorf, J., Juma, S., et al., 2019c. Evaluation of integrated interventions layered on mass drug administration for urogenital schistosomiasis elimination: a cluster-randomised trial. Lancet Glob. Health 7, e1118–e1129.

Knopp, S., Person, B., Ame, S.M., Ali, S.M., Mohammed, K.A., Rollinson, D., 2015b. Elimination of urogenital schistosomiasis in Zanzibar: Challenges for an integrated multidisciplinary research programme. Trop. Med. Int. Health 20, 230–231.

Knopp, S., Person, B., Ame, S.M., Ali, S.M., Muhsin, J., Juma, S., et al., 2016. Praziquantel coverage in schools and communities targeted for the elimination of urogenital schistosomiasis in Zanzibar: a cross-sectional survey. Parasit. Vectors 9, 5.

Knopp, S., Person, B., Ame, S.M., Mohammed, K.A., Ali, S.M., Khamis, I.S., et al., 2013a. Elimination of schistosomiasis transmission in Zanzibar: Baseline findings before the onset of a randomized intervention trial. PLOS Negl. Trop. Dis. 7, e2474.

Knopp, S., Stothard, J.R., Rollinson, D., Mohammed, K.A., Khamis, I.S., Marti, H., et al., 2013b. From morbidity control to transmission control: Time to change tactics against helminths on Unguja Island, Zanzibar. Acta Trop. 128, 412–422.

Kura, K., Ayabina, D., Hollingsworth, T.D., Anderson, R.M., 2022. Determining the optimal strategies to achieve elimination of transmission for *Schistosoma mansoni*. Parasit. Vectors 15, 55.

Le Clec'h, W., Chevalier, F.D., Mattos, A.C.A., Strickland, A., Diaz, R., Mcdew-White, M., et al., 2021. Genetic analysis of praziquantel response in schistosome parasites implicates a transient receptor potential channel. Sci. Transl. Med. 13, eabj9114.

Léger, E., Garba, A., Hamidou, A.A., Webster, B.L., Pennance, T., Rollinson, D., et al., 2016. Introgressed animal schistosomes *Schistosoma curassoni* and *S. bovis* naturally infecting humans. Emerg. Infect. Dis. 22, 2212–2214.

Levecke, B., Vlaminck, J., Andriamaro, L., Ame, S., Belizario, V., Degarege, A., et al., 2020. Evaluation of the therapeutic efficacy of praziquantel against schistosomes in seven countries with ongoing large-scale deworming programs. Int. J. Parasitol.: Drugs Drug. Resist. 14, 183–187.

Lund, A.J., Wade, K.J., Nikolakis, Z.L., Ivey, K.N., Perry, B.W., Pike, H.N.C., et al., 2022. Integrating genomic and epidemiologic data to accelerate progress toward schistosomiasis elimination. eLife 11, e79320.

Lwambo, N.J.S., Savioli, L., Kisumku, U.M., Alawi, K.S., Bundy, D.A.P., 1997a. Control of *Schistosoma haematobium* morbidity on Pemba Island: Validity and efficiency of indirect screening tests. Bull. World Health Organ. 75, 247–252.

Lwambo, N.J.S., Savioli, L., Kisumku, U.M., Alawi, K.S., Bundy, D.A.P., 1997b. The relationship between prevalence of *Schistosoma haematobium* infection and different

morbidity indicators during the course of a control programme on Pemba Island. Trans. R. Soc. Trop. Med. Hyg. 91, 643–646.
Lyons, B., Stothard, R., Rollinson, D., Khamis, S., Simai, K.A., Hunter, P.R., 2009. A comparison of urinary tract pathology and morbidity in adult populations from endemic and non-endemic zones for urinary schistosomiasis on Unguja Island, Zanzibar. BMC Infect. Dis. 9, 189.
Macdonald, G., Forsyth, D.M., 1968. Urological complications of endemic schistosomiasis in schoolchildren Part 3. Follow-up studies at Donge School, Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 62, 766–774.
Macdonald, G., Forsyth, D.M., Rashid, C., 1968a. Urological complications of endemic schistosomiasis in schoolchildren Part 4. As modified by treatment. Trans. R. Soc. Trop. Med. Hyg. 62, 775–781.
Macdonald, G., Forsyth, D.M., Rayski, C., Rashid, C., 1968b. The control of urinary schistosomiasis. Trials of niridazole and other drugs in children attending primary schools in Zanzibar, Tanzania, and the failure of a pilot eradication programme. Ann. Trop. Med. Parasitol. 62, 407–415.
Maier, T., Wheeler, N.J., Namigai, E.K.O., Tycko, J., Grewelle, R.E., Woldeamanuel, Y., et al., 2019. Gene drives for schistosomiasis transmission control. PLOS Negl. Trop. Dis. 13, e0007833.
Mandahl-Barth, G., 1957. Intermediate hosts of *Schistosoma*; *African Biomphalaria* and *Bulinus*. I. Bull. World Health Organ. 16, 1103–1163.
Mandahl-Barth, G., 1958. Intermediate hosts of *Schistosoma*; *African Biomphalaria* and *Bulinus*. Monograph Series World Health Organization, pp. 1–131.
Mandahl-Barth, G., 1963. WHO International Snail Identification Centre Charlottenlund - Rep. 1963. *From* WHO Archives B2_181_4. 32–33.
Manning, W.R., 1976. Zanzibar: Schistosomiasis Control Project. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 487–503.
Mansfield-Aders, W., 1928. Annual Report of the Biological Division for 1927. *From* Wellcome Collect.
Mazigo, H.D., Samson, A., Lambert, V.J., Kosia, A.L., Ngoma, D.D., Murphy, R., et al., 2021. we know about schistosomiasis but we know nothing about FGS: A qualitative assessment of knowledge gaps about female genital schistosomiasis among communities living in *Schistosoma haematobium* endemic districts of Zanzibar and Northwestern Tanzania. PLOS Negl. Trop. Dis. 15.
Mccarthy, D.D., 1930. Medical notes from Weti, Pemba. Trans. R. Soc. Trop. Med. Hyg. 13, 401–412.
Mccullough, F.S., 1978. Organization of Schistosomiasis Control in the African Region. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 351–387.
Mccullough, F.S., Hsueh, C.-I., Tembo, D., Ahmed, H.T., Mwita, U., 1979. Interim Plan of Action for an Operational Research Study of the Delivery of Feasible Schistosomiasis Control Methods in Zanzibar, Utilizing Primary Health Care Approaches and Community Participation - Report on a Site Visit from 11 to 16 August, 1979. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 326–350.
Mccullough, F.S., Krafft, J.G., 1976. Schistosomiasis in Zanzibar and Pemba Tanzan. - Rep. a Mission. 1 April. - 7 June 1975. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 430–486.
Mesquita, S.G., Lugli, E.B., Matera, G., Fonseca, C.T., Caldeira, R.L., Webster, B., 2022. Development of real-time and lateral flow recombinase polymerase amplification assays for rapid detection of *Schistosoma mansoni*. Front. Microbiol. 13, 1043596.
Mgeni, A.F., 1984a. Proposal Document for the Control of Schistosomiasis in Zanzibar Islands. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 596–601.

Mgeni, A.F., 1984b. Extension Programme for the Control of Schistosomiasis in Zanzibar. *From* WHO Archives.
Mgeni, A.F., 1995a. Collaborative Research Project - Section B: Proposal Form. *From* WHO Archives 1995a 950780.
Mgeni, A.F., 1995b. First School Deworming Programme from 13/395 to 13/7/1995. *From* WHO Archives P2/370/4ZAN.
Mgeni, A.F., 2002. Progress Reports, Final Reports & Renewal Requests - Section B: Form. *From* WHO Archives.
Mgeni, A.F. n.d. Control of Urinary Schistosomiasis under Primary Health Care Approach in Zanzibar. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 504–530.
Mgeni, A.F., Kisumku, U.M., Mccullough, F.S., Dixon, H., Yoon, S.S., Mott, K.E., 1990. Metrifonate in the control of urinary schistosomiasis in Zanzibar. Bull. World Health Organ. 68, 721–730.
Ministry of Finance and Planning - National Bureau of Statistics Tanzania & President's Office - Finance and Planning - Office of the Chief Government Statistician Zanzibar, 2022. Administrative Units Population Distribution Report. *From* ⟨www.nbs.go.tz/index.php/en/⟩.
Ministry of Health (Moh) [Tanzania Mainland], Ministry of Health (MOH) [Zanzibar], National Bureau of Statistics (Nbs), Office of the Chief Government Statistician (OCGS) & ICF, 2023. Tanzania Demographic and Health Survey and Malaria Indicator Survey 2022 Key Indicators Report. *From* Dodoma, Tanzania, and Rockville, Maryland, USA.
Mkoji, G.M., Hofkin, B.V., Kuris, A.M., Stewart-Oaten, A., Mungai, B.N., Kihara, J.H., et al., 1999. Impact of the crayfish *Procambarus clarkii* on *Schistosoma haematobium* transmission in Kenya. Am. J. Trop. Med. Hyg. 61, 751–759.
Mohammed, K.A., Haji, H.J., Gabrielli, A.F., Mubila, L., Biswas, G., Chitsulo, L., et al., 2008. Triple co-administration of ivermectin, albendazole and praziquantel in Zanzibar: a safety study. PLOS Negl. Trop. Dis. 2, e171.
Mohammed, K.A., Molyneux, D.H., Albonico, M., Rio, F., 2006. Progress towards eliminating lymphatic filariasis in Zanzibar: a model programme. Trends Parasitol. 22, 340–344.
Montresor, A., Crompton, D.W.T., Gyorkos, T.W., Savioli, L., World Health Organization, 2002. Helminth Control in School-Age Children: A Guide for Managers of Control Programmes. World Health Organization, Geneva.
Montresor, A., Engels, D., Ramsan, M., Foum, A., Savioli, L., 2002b. Field test of the 'dose pole' for praziquantel in Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 96, 323–324.
Montresor, A., Odermatt, P., Muth, S., Iwata, F., Raja'a, Y.A., Assis, A.M., et al., 2005. The WHO dose pole for the administration of praziquantel is also accurate in non-African populations. Trans. R. Soc. Trop. Med. Hyg. 99, 78–81.
Montresor, A., Ramsan, M., Chwaya, H.M., Ameir, H., Foum, A., Albonico, M., et al., 2001. Extending anthelminthic coverage to non-enrolled school-age children using a simple and low-cost method. Trop. Med. Int. Health 6, 535–537.
Mott, K.E., Mccullough, F.S., Dixon, H., 1983. Duty Travel Report - United Republic of Tanzania - 28 February - 5 March 1983. *From* WHO Archives WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 602–626.
Mozley, A., 1939. XXVI.—The fresh-water mollusca of the Tanganyika territory and Zanzibar Protectorate, and their relation to human schistosomiasis. Trans. R. Soc. Edinb. 59, 687–744.
Muchiri, E.M., 1991. Report on Visit to Pemba (Schistosomiasis control program); 19 November to 1 December 1991. *From* WHO Archives B2_370_3TAN.
Muhammed, M., 1980. The Prevalence of Urinary Schistosomiasis as Related to Source of Water Supply and Some Personal Habits in Schoolchildren in a Rural Community at

Donge, Zanzibar, Tanzania. *From* WHO Archives WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 294–310.
Mujumbusi, L., Nalwadda, E., Ssali, A., Pickering, L., Seeley, J., Meginnis, K., et al., 2023. Understanding perceptions of schistosomiasis and its control among highly endemic lakeshore communities in Mayuge, Uganda. PLOS Negl. Trop. Dis. 17, e0010687.
Mutani, A., Christensen, N.O., Frandsen, F., 1983. Studies on the relationship between *Schistosoma* and their intermediate hosts. V. The genus *Bulinus* and *Schistosoma bovis* from Iringa, Tanzania. Z. Parasitenkd. 69, 483–487.
Neves, M.I., Gower, C.M., Webster, J.P., Walker, M., 2021. Revisiting density-dependent fecundity in schistosomes using sibship reconstruction. PLOS Negl. Trop. Dis. 15, e0009396.
Office of Chief Government Statistician Zanzibar, 2020. Zanzibar in Figures 2019 [Online]. Available: ⟨https://www.ocgs.go.tz/⟩ [Accessed].
Office of Chief Government Statistician Zanzibar & Ministry of Finance and Planning Zanzibar, 2020. 2019/20 Household Budget Survey [Online]. Available: https://www.ocgs.go.tz/ [Accessed].
Page, M.J., Mckenzie, J.E., Bossuyt, P.M., Boutron, I., Hoffmann, T.C., Mulrow, C.D., et al., 2021. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. BMJ 372, n71.
Pampiglione, S., Visconti, S., Stefanini, A., 1987. Human intestinal parasites in Subsaharan Africa. III. Pemba Island (Zanzibar-Tanzania). Parassitologia 29, 27–35.
Park, S.-K., Gunaratne, G.S., Chulkov, E.G., Moehring, F., Mccusker, P., Dosa, P.I., et al., 2019. The anthelmintic drug praziquantel activates a schistosome transient receptor potential channel. J. Biol. Chem. 294, 18873–18880.
Pearson, M.S., Tedla, B.A., Mekonnen, G.G., Proietti, C., Becker, L., Nakajima, R., et al., 2021. Immunomics-guided discovery of serum and urine antibodies for diagnosing urogenital schistosomiasis: a biomarker identification study. Lancet Microbe 2, e617–e626.
Pellegrino, J., 1958. The intradermal test in the diagnosis of bilharziasis. Bull. World Health Organ. 18, 945–961.
Pennance, T. 2020. Genetic Diversity and Evolution within the Genus *Bulinus* and Species-level Interactions with the Transmission of *Schistosoma haematobium* Group Parasites.
Pennance, T., Allan, F., Emery, A., Rabone, M., Cable, J., Garba, A.D., et al., 2020a. Interactions between *Schistosoma haematobium* group species and their *Bulinus* spp. intermediate hosts along the Niger River Valley. Parasit. Vectors 13, 268.
Pennance, T., Ame, S.M., Amour, A.K., Suleiman, K.R., Allan, F., Rollinson, D., et al., 2018. Corrigendum: Occurrence of *Schistosoma bovis* on Pemba Island, Zanzibar: Implications for urogenital schistosomiasis transmission monitoring. Parasitology 145, 1732. https://doi.org/10.1017/S0031182018001154
Pennance, T., Ame, S.M., Amour, A.K., Suleiman, K.R., Cable, J., Webster, B.L., 2021. The detection of *Schistosoma bovis* in livestock on Pemba Island, Zanzibar: A preliminary study. Curr. Res. Parasitol. Vector-Borne Dis. 1, 100056.
Pennance, T., Ame, S.M., Amour, A.K., Suleiman, K.R., Muhsin, M.A., Kabole, F., et al., 2022a. Transmission and diversity of *Schistosoma haematobium* and *S. bovis* and their freshwater intermediate snail hosts *Bulinus globosus* and *B. nasutus* in the Zanzibar Archipelago, United Republic of Tanzania. PLOS Negl. Trop. Dis. 16, e0010585.
Pennance, T., Archer, J., Lugli, E.B., Rostron, P., Llanwarne, F., Ali, S.M., et al., 2020b. Development of a molecular snail xenomonitoring assay to detect *Schistosoma haematobium* and *Schistosoma bovis* infections in their *Bulinus* snail hosts. Molecules (Basel, Switz.) 25, 4011.
Pennance, T., Neves, M.I., Webster, B.L., Gower, C.M., Knopp, S., Khamis, I.S., et al., 2022b. Potential Drivers for Schistosomiasis Persistence: Population Genetic Analyses

from a Cluster-randomized Urogenital Schistosomiasis Elimination Trial across the Zanzibar Islands.
Pennance, T., Person, B., Muhsin, M.A., Khamis, A.N., Muhsin, J., Khamis, I.S., et al., 2016. Urogenital schistosomiasis transmission on Unguja Island: Characterisation of persistent hot-spots, Zanzibar. Parasit. Vectors 9, 646.
Pereira, L.P.L.A., Ribeiro, E.C.G., Brito, M.C.A., Silveira, D.P.B., Araruna, F.O.S., Araruna, F.B., et al., 2020. Essential oils as molluscicidal agents against schistosomiasis transmitting snails - a review. Acta Trop. 209, 105489.
Person, B., Ali, S.M., A'kadir, F.M., Ali, J.N., Mohammed, U.A., Mohammed, K.A., Rollinson, D., et al., 2016a. Community knowledge, perceptions, and practices associated with urogenital schistosomiasis among school-aged children in Zanzibar, United Republic of Tanzania. PLOS Negl. Trop. Dis. 10, e0004814.
Person, B., Knopp, S., Ali, S.M., A'kadir, F.M., Khamis, A.N., Ali, J.N., et al., 2016b. Community co-designed schistosomiasis control interventions for school-aged children in Zanzibar. J. Biosoc. Sci. 48, S56–S73.
Person, B., Mohammed, K., Ali, S., Rollinson, D., Knopp, S., 2012. Community perceptions of schistosomiasis risk among school children in Zanzibar. Am. J. Trop. Med. Hyg. 87, 438.
Person, B., Rollinson, D., Ali, S.M., Mohammed, U.A., A'kadir, F.M., Kabole, F., et al., 2021. Evaluation of a urogenital schistosomiasis behavioural intervention among students from rural schools in Unguja and Pemba islands, Zanzibar. Acta Trop. 220, 105960.
Platt, R.N., Mcdew-White, M., Clec'h, L.E., Chevalier, W., Allan, F.D., Emery, F., et al., 2019. Ancient hybridization and adaptive introgression of an invadolysin gene in schistosome parasites. Mol. Biol. Evol. 36, 2127–2142.
Portnoy, J., Brewer, J.H., Harris, A., 1962. Rapid plasma reagin card test for syphilis and other treponematoses. Public Health Rep. (1896) 77, 645–652.
Prendergast, M.E., Rouby, H., Punnwong, P., Marchant, R., Crowther, A., Kourampas, N., et al., 2016. Continental island formation and the archaeology of defaunation on Zanzibar, eastern Africa. PLOS One 11, e0149565.
Rebollo, M.P., Mohammed, K.A., Thomas, B., Ame, S., Ali, S.M., Cano, J., et al., 2015. Cessation of mass drug administration for lymphatic filariasis in Zanzibar in 2006: was transmission interrupted? PLOS Negl. Trop. Dis. 9, e0003669.
Rey, O., Webster, B.L., Huyse, T., Rollinson, D., Den Broeck, V.A.N., Kincaid-Smith, F., et al., 2021. Population genetics of African *Schistosoma* species. Infect. Genet. Evol. 89, 104727.
Rollinson, D., Klinger, E.V., Mgeni, A.F., Khamis, I.S., Stothard, J.R., 2005. Urinary schistosomiasis on Zanzibar: Application of two novel assays for the detection of excreted albumin and haemoglobin in urine. J. Helminthol. 79, 199–206.
Rollinson, D., Southgate, V.R., 1979. Enzyme analyses of *Bulinus africanus* group snails (Mollusca: Planorbidae) from Tanzania. Trans. R. Soc. Trop. Med. Hyg. 73, 667–672.
Rollinson, D., Webster, B., Diaw, O., Kabatereine, N., Khamis, I.S., Stothard, R., 2009a. A new molecular epidemiology of African schistosomiasis with focus upon schistosome diversity. Trop. Med. Int. Health 14, 8.
Rollinson, D., Webster, J.P., Webster, B., Nyakaana, S., Jørgensen, A., Stothard, J.R., 2009b. Genetic diversity of schistosomes and snails: implications for control. Parasitology 136, 1801–1811.
Rollinson, D., Wright, C.A., 1984. Population studies on *Bulinus cernicus* from Mauritius. Malacologia 25, 447–463.
Ross, A.G.P., Olveda, R.M., Chy, D., Olveda, D.U., Li, Y., Harn, D.A., et al., 2014. Can mass drug administration lead to the sustainable control of schistosomiasis? J. Infect. Dis. 211, 283–289.

Rostron, P., Pennance, T., Bakar, F., Rollinson, D., Knopp, S., Allan, F., et al., 2019. Development of a recombinase polymerase amplification (RPA) fluorescence assay for the detection of *Schistosoma haematobium*. Parasit. Vectors 12, 514.
Rowson, B., Warren, B., Ngereza, C.F., 2010. Terrestrial molluscs of Pemba Island, Zanzibar, Tanzania, and its status as an "oceanic" island. ZooKeys (70), 1–39.
Rudge, J.W., Stothard, J.R., Basanez, M.G., Mgeni, A.F., Khamis, I.S., Khamis, A.N., et al., 2008. Micro-epidemiology of urinary schistosomiasis in Zanzibar: Local risk factors associated with distribution of infections among schoolchildren and relevance for control. Acta Trop. 105, 45–54.
Sadun, E.H., Anderson, R.I., Schoenbechler, M.J., 1963. A plasma card test for rapid serodiagnosis of schistosomiasis (SPC). Proc. Soc. Exp. Biol. Med. 112, 280–283.
Sady, H., Al-Mekhlafi, H.M., Webster, B.L., Ngui, R., Atroosh, W.M., Al-Delaimy, A.K., et al., 2015. New insights into the genetic diversity of *Schistosoma mansoni* and *S. haematobium* in Yemen. Parasit. Vectors 8, 544.
Sakho, F., Badila, C.F., Dembele, B., Diaby, A., Camara, A.K., Lamah, L., et al., 2021. Implementation of mass drug administration for neglected tropical diseases in Guinea during the COVID-19 pandemic. PLOS Negl. Trop. Dis. 15, e0009807.
Salari, P., Fürst, T., Knopp, S., Rollinson, D., Kabole, F., Khamis, M.I., et al., 2020. Financial costs of the Zanzibar Elimination of Schistosomiasis Transmission project. Am. J. Trop. Med. Hyg. 103, 2260–2267.
Savioli, L., 1991a. The control of bladder schistosomiasis on the isle of Pemba. A balance after the first five years (1986-1991). Pharm. Ind. 53, 1009–1012.
Savioli, L., 1991b. Travel Report Summary. *From* WHO WHO007_SCHISTO1_AFRO_ZANZIBAR_1. 535 537.
Savioli, L., 1994. Duty Travel Report Germany and United Republic of Tanzania 10/03 - 03/04/94. *From* WHO Archives.
Savioli, L., 1995. Travel Report Summary. *From* WHO Archives P2_370_4ZAN.
Savioli, L., Albonico, M., Amour, K.A., Saleh, J.M., 1993. Expired reagent strips for haematuria. Trop. Dr. 23, 187–188.
Savioli, L., Albonico, M., Engels, D., Montresor, A., 2004. Progress in the prevention and control of schistosomiasis and soil-transmitted helminthiasis. Parasitol. Int. 53, 103–113.
Savioli, L., Dixon, H., Kisumku, U.M., Mott, K.E., 1989a. Control of morbidity due to *Schistosoma haematobium* on Pemba island: Programme organization and management. Trop.Med. Parasitol. 40, 189–194.
Savioli, L., Dixon, H., Kisumku, U.M., Mott, K.E., 1989b. Control of morbidity due to *Schistosoma haematobium* on Pemba Island; selective population chemotherapy of schoolchildren with haematuria to identify high-risk localities. Trans. R. Soc. Trop. Med. Hyg. 83, 805–810.
Savioli, L., Gabrielli, A., Neve, H., 1990a. Vulvar *Schistosoma haematobium* lesion treated with praziquantel. Trop. Dr. 20, 45–46.
Savioli, L., Hatz, C., Dixon, H., Kisumku, U.M., Mott, K.E., 1990b. Control of morbidity due to *Schistosoma haematobium* on Pemba Island: Egg excretion and hematuria as indicators of infection. Am. J. Trop. Med. Hyg. 43, 289–295.
Savioli, L., Mott, K.E., 1989. Urinary schistosomiasis on Pemba Island: Low-cost diagnosis for control in a primary health care setting. Parasitol. Today 5, 333–337.
Scientific Working Group on Schistosomiasis, Meeting, UNICEF/UNDP/WORLD BANK/WHO Special Programme For Research, Training In Tropical Diseases & World Health Organization, 2006. Report of the Scientific Working Group meeting on Schistosomiasis, Geneva, 14–16 November, 2005. World Health Organization, Geneva.
Semple, J.M., 1927. Public Health. *From* Wellcome Collect.

Sengupta, M.E., Hellström, M., Kariuki, H.C., Olsen, A., Thomsen, P.F., Mejer, H., et al., 2019. Environmental DNA for improved detection and environmental surveillance of schistosomiasis. Proc. Natl. Acad. Sci. 116, 8931–8940.

Shauri, O.M., 1994. Support to the Construction of the Pemba Public Health Laboratory. *From* WHO Archives P2_370_4ZAN 1994.

Shortt, J.A., Timm, L.E., Hales, N.R., Nikolakis, Z.L., Schield, D.R., Perry, B.W., et al., 2021. Population genomic analyses of schistosome parasites highlight critical challenges facing endgame elimination efforts. Sci. Rep. 11, 6884.

Simonová, J., Simon, O.P., Kapic, Š., Nehasil, L., Horsák, M., 2016. Medium-sized forest snails survive passage through birds' digestive tract and adhere strongly to birds' legs: more evidence for passive dispersal mechanisms. J. Molluscan Stud. 82, 422–426.

Sokolow, S.H., Huttinger, E., Jouanard, N., Hsieh, M.H., Lafferty, K.D., Kuris, A.M., et al., 2015. Reduced transmission of human schistosomiasis after restoration of a native river prawn that preys on the snail intermediate host. Proc. Natl. Acad. Sci. U. S. A. 112, 9650–9655.

Sousa-Figueiredo, J.C., Basáñez, M.G., Khamis, I.S., Garba, A., Rollinson, D., Stothard, J.R., 2009. Measuring morbidity associated with urinary schistosomiasis: assessing levels of excreted urine albumin and urinary tract pathologies. PLOS Negl. Trop. Dis. 3, e526.

Sousa-Figueiredo, J.C., Basáñez, M.G., Mgeni, A.F., Khamis, I.S., Rollinson, D., Stothard, J.R., 2008. A parasitological survey, in rural Zanzibar, of pre-school children and their mothers for urinary schistosomiasis, soil-transmitted helminthiases and malaria, with observations on the prevalence of anaemia. Ann. Trop. Med. Parasitol. 102, 679–692.

Sousa-Figueiredo, J.C., Betson, M., Garba, A., Sacko, M., Vaz-Nery, S., Amin, M.A., et al., 2011. Treatment of schistosomiasis in infants and preschool-children with Praziquantel: correct dose, side-effects and cure rates. Am. J. Trop. Med. Hyg. 85, 143.

Sousa-Figueiredo, J.C., Betson, M., Stothard, J.R., 2012. Treatment of schistosomiasis in African infants and preschool-aged children: downward extension and biometric optimization of the current praziquantel dose pole. Int. Health 4, 95–102.

Southgate, V.R., Rollinson, D., Tchuem Tchuenté, L.A., Hagan, P., 2005. Towards control of schistosomiasis in sub-Saharan Africa. J. Helminthol. 79, 181–185.

Spaan, J.M., Pennance, T., Laidemitt, M.R., Sims, N., Roth, J., Lam, Y., et al., 2023. Multi-strain compatibility polymorphism between a parasite and its snail host, a neglected vector of schistosomiasis in Africa. Curr. Res. Parasitol. Vector-Borne Dis. 3, 100120.

Stoltzfus, R.J., Albonico, M., Tielsch, J.M., Chwaya, H.M., Savioli, L., 1997a. Linear growth retardation in Zanzibari school children. J. Nutr. 127, 1099–1105.

Stoltzfus, R.J., Chwaya, H.M., Tielsch, J.M., Schulze, K.J., Albonico, M., Savioli, L., 1997b. Epidemiology of iron deficiency anemia in Zanzibari schoolchildren: the importance of hookworms. Am. J. Clin. Nutr. 65, 153–159.

Stothard, J.R., Ameri, H., Khamis, I.S., Blair, L., Nyandindi, U.S., Kane, R.A., et al., 2013. Parasitological and malacological surveys reveal urogenital schistosomiasis on Mafia Island, Tanzania to be an imported infection. Acta Trop. 128, 326–333.

Stothard, J.R., Brémond, P., Andriamaro, L., Sellin, B., Sellin, E., Rollinson, D., 2001. *Bulinus* species on Madagascar: molecular evolution, genetic markers and compatibility with *Schistosoma haematobium*. Parasitology 123 (Suppl), S261–S275.

Stothard, J.R., French, M.D., Khamis, I.S., Basáñez, M.G., Rollinson, D., 2009a. The epidemiology and control of urinary schistosomiasis and soil-transmitted helminthiasis in schoolchildren on Unguja Island, Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 103, 1031–1044.

Stothard, J.R., Kabatereine, N.B., Tukahebwa, E.M., Kazibwe, F., Rollinson, D., Mathieson, W., et al., 2006a. Use of circulating cathodic antigen (CCA) dipsticks for detection of intestinal and urinary schistosomiasis. Acta Trop. 97, 219–228.

Stothard, J.R., Khamis, A.N., Khamis, I.S., Neo, C.H.E., Wei, I., Rollinson, D., 2016. Health education and the control of urogenital schistosomiasis: assessing the impact of the Juma Na Kichocho comic-strip medical booklet in Zanzibar. J. Biosoc. Sci. 48, S40–S55.
Stothard, J.R., Llewellyn-Hughes, J., Griffin, C.E., Hubbard, S.J., Kristensen, T.K., Rollinson, D., 2002a. Identification of snails within the Bulinus africanus group from East Africa by multiplex SnaPshot™ analysis of single nucleotide polymorphisms within the cytochrome oxidase subunit I. Mem. Inst. Oswaldo Cruz 97, 31–36.
Stothard, J.R., Loxton, N., Rollinson, D., Mgeni, A.F., Khamis, S., Ameri, H., et al., 2000. The transmission status of *Bulinus* on Zanzibar Island (Unguja), with implications for control of urinary schistosomiasis. Ann. Trop. Med. Parasitol. 94, 87–94.
Stothard, J.R., Loxton, N.J., Rollinson, D., 2002b. Freshwater snails on Mafia Island, Tanzania with special emphasis upon the genus *Bulinus* (Gastropoda: Planorbidae). J. Zool. 257, 353–364.
Stothard, J.R., Mgeni, A.F., Alawi, K.S., Savioli, L., Rollinson, D., 1997. Observations on shell morphology, enzymes and random amplified polymorphic DNA (RAPD) in *Bulinus Africanus* group snails (Gastropoda: Planorbidae) in Zanzibar. J. Molluscan Stud. 63, 489–503.
Stothard, J.R., Mgeni, A.F., Khamis, S., Seto, E., Ramsan, M., Hubbard, S.J., et al., 2002c. Fresh from the fields. New Insights into the transmission biology of urinary schistosomiasis in Zanzibar. Trans. R. Soc. Trop. Med. Hyg. 96, 470–475.
Stothard, J.R., Mgeni, A.F., Khamis, S., Seto, E., Ramsan, M., Rollinson, D., 2002d. Urinary schistosomiasis in schoolchildren on Zanzibar Island (Unguja), Tanzania: a parasitological survey supplemented with questionnaires. Trans. R. Soc. Trop. Med. Hyg. 96, 507–514.
Stothard, J.R., Mook, P., Mgeni, A.F., Khamis, I.S., Khamis, A.N., Rollinson, D., 2006b. Control of urinary schistosomiasis on Zanzibar (Unguja Island): a pilot evaluation of the educational impact of the Juma na Kichocho health booklet within primary schools. Mem. Inst. Oswaldo Cruz 101, 119–124.
Stothard, J.R., Rollinson, D., 1997a. Partial DNA sequences from the mitochondrial cytochrome oxidase subunit I (COI) gene can differentiate the intermediate snail hosts *Bulinus globosus* and *B. nasutus* (Gastropoda: Planorbidae). J. Nat. Hist. 31, 727–737.
Stothard, J.R., Rollinson, D., 1997b. Molecular characterization of *Bulinus globosus* and *B. nasutus* on Zanzibar, and an investigation of their roles in the epidemiology of *Schistosoma haematobium*. Trans. R. Soc. Trop. Med. Hyg. 91, 353–357.
Stothard, J.R., Sousa-Figueiredo, J.C., Khamis, I.S., Garba, A., Rollinson, D., 2009b. Urinary schistosomiasis-associated morbidity in schoolchildren detected with urine albumin-to-creatinine ratio (UACR) reagent strips. J. Pediatr. Urol. 5, 287–291.
Stothard, J.R., Sousa-Figueiredo, J.C., Standley, C., Dam, V.A.N., Knopp, G.J., Utzinger, S., et al., 2009c. An evaluation of urine-CCA strip test and fingerprick blood SEA-ELISA for detection of urinary schistosomiasis in schoolchildren in Zanzibar. Acta Trop. 111, 64–70.
Sturrock, R.F., 1965. The development of irrigation and its influence on the transmission of bilharziasis in Tanganyika. Bull. World Health Organ. 32, 225–236.
Sturrock, R.F., 1966. The influence of temperature on the biology of *Biomphalaria pfeifferi* (Krauss), an intermediate host of *Schistosoma mansoni*. Ann. Trop. Med. Parasitol. 60, 100–105.
Sturt, A.S., Webb, E.L., Francis, S.C., Hayes, R.J., Bustinduy, A.L., 2020. Beyond the barrier: female genital schistosomiasis as a potential risk factor for HIV-1 acquisition. Acta Trop. 209, 105524.
Sullivan, J.T., Cheng, T.C., Chen, C.C., 1984. Genetic selection for tolerance to niclosamide and copper in *Biomphalara glabrata* (Mollusca: Pulmonata). Z. Tropenmed. Parasitol. 35, 189–192.

Tembo, R., Muleya, W., Yabe, J., Kainga, H., Nalubamba, K.S., Zulu, M., et al., 2022. Prevalence and molecular identification of Schistosoma haematobium among children in Lusaka and Siavonga districts, Zambia. Trop. Med. Infect. Dis. 7, 239.

Tennessen, J.A., Bollmann, S.R., Peremyslova, E., Kronmiller, B.A., Sergi, C., Hamali, B., et al., 2020. Clusters of polymorphic transmembrane genes control resistance to schistosomes in snail vectors. eLife 9, e59395.

1Trippler, L., Ali, M.N., Ame, S.M., Ali, S.M., Kabole, F., Hattendorf, J., et al., 2022a. GPS-based fine-scale mapping surveys for schistosomiasis assessment: a practical introduction and documentation of field implementation. Infect. Dis. Poverty 11, 8.

Trippler, L., Ali, S.M., Ame, S.M., Hattendorf, J., Suleiman, K.R., Ali, M.N., et al., 2022b. Fine-scale-mapping of *Schistosoma haematobium* infections at the school and community levels and intermediate host snail abundance in the north of Pemba Island: baseline cross-sectional survey findings before the onset of a 3-year intervention study. Parasit. Vectors 15, 292.

Trippler, L., Ame, S.M., Hattendorf, J., Juma, S., Abubakar, S., Ali, S.M., et al., 2021a. Impact of seven years of mass drug administration and recrudescence of *Schistosoma haematobium* infections after one year of treatment gap in Zanzibar: repeated cross-sectional studies. PLOS Negl. Trop. Dis. 15, 1–21.

Trippler, L., Hattendorf, J., Ali, S.M., Ame, S.M., Juma, S., Kabole, F., et al., 2021b. Novel tools and strategies for breaking schistosomiasis transmission: study protocol for an intervention study. BMC Infect. Dis. 21, 1024.

Tschuem Tchuenté, L.A., N'goran, E.K., 2009. Schistosomiasis and soil-transmitted helminthiasis control in Cameroon and Côte d'Ivoire: implementing control on a limited budget. Parasitology 136, 1739–1745.

United Nations. Geospatial Information Section, 1987. Hydrogeological Map of Zanzibar Including the Islands of Zanzibar and Pemba, the United Republic of Tanzania [cartographic material]. Downloaded on 04.05.2023.

Utzinger, J., Becker, S.L., Van Lieshout, L., Van Dam, G.J., Knopp, S., 2015. New diagnostic tools in schistosomiasis. Clin. Microbiol. Infect. 21, 529–542.

Van Dam, G.J., Wichers, J.H., Ferreira, T.M., Ghati, D., Van Amerongen, A., Deelder, A.M., 2004. Diagnosis of schistosomiasis by reagent strip test for detection of circulating cathodic antigen. J. Clin. Microbiol. 42, 5458–5461.

Van Etten, L., Folman, C.C., Eggelte, T.A., Kremsner, P.G., Deelder, A.M., 1994. Rapid diagnosis of schistosomiasis by antigen detection in urine with a reagent strip. J. Clin. Microbiol. 32, 2404–2406.

Vreysen, M.J.B., Saleh, K., Ali, M., Abdulla, A., Zhu, Z.-R., Juma, K., 2000. The use of the sterile insect technique (SIT) for the eradication of the tsetse fly Glossina austeni (Diptera: Glossinidae) on the Island of Unguja (Zanzibar). J. Econ. Entomol. 93, 123–135.

Wang, X.Y., He, J., Juma, S., Kabole, F., Guo, J.G., Dai, J.R., et al., 2019. Efficacy of China-made praziquantel for treatment of *Schistosomiasis haematobium* in Africa: a randomized controlled trial. PLoS Negl. Trop. Dis. 13, e0007238.

Webster, B.L., Culverwell, C.L., Khamis, I.S., Mohammed, K.A., Rollinson, D., Stothard, J.R., 2013a. DNA barcoding of *Schistosoma haematobium* on Zanzibar reveals substantial genetic diversity and two major phylogenetic groups. Acta Trop. 128, 206–217.

Webster, B.L., Diaw, O.T., Seye, M.M., Webster, J.P., Rollinson, D., 2013b. Introgressive hybridization of *Schistosoma haematobium* group species in Senegal: species barrier break down between ruminant and human schistosomes. PLOS Negl. Trop. Dis. 7, e2110.

Webster, B.L., Emery, A.M., Webster, J.P., Gouvras, A., Garba, A., Diaw, O., et al., 2012. Genetic diversity within *Schistosoma haematobium*: DNA barcoding reveals two distinct groups. PLOS Negl. Trop. Dis. 6, e1882.

Webster, B.L., Rabone, M., Pennance, T., Emery, A.M., Allan, F., Gouvras, A., et al., 2015. Development of novel multiplex microsatellite polymerase chain reactions to

enable high-throughput population genetic studies of *Schistosoma haematobium*. Parasites & Vectors 8, 432.
Webster, B.L., Rollinson, D., Stothard, J.R., Huyse, T., 2010. Rapid diagnostic multiplex PCR (RD-PCR) to discriminate *Schistosoma haematobium* and *S. bovis*. J. Helminthol. 84, 107–114.
Webster, B.L., Southgate, V.R., Littlewood, D.T., 2006. A revision of the interrelationships of *Schistosoma* including the recently described *Schistosoma guineensis*. Int. J. Parasitol. 36, 947–955.
Webster, J.P., Neves, M.I., Webster, B.L., Pennance, T., Rabone, M., Gouvras, A.N., et al., 2020. Parasite population genetic contributions to the schistosomiasis consortium for operational research and evaluation within sub-Saharan Africa. Am. J. Trop. Med. Hyg. 103, 80–91.
Weerakoon, K.G., Gobert, G.N., Cai, P., Mcmanus, D.P., 2015. Advances in the diagnosis of human schistosomiasis. Clin. Microbiol. Rev. 28, 939–967.
WHO Archives, 1955–1960. Correspondence. *From* WHO Archives B2_374_3.
WHO Archives, n.d. Schistosomiasis Control in Zanzibar. *From* WHO Archives WHO007_SCHISTO_1_AFRO_ZANZIBAR_1. 2–9.
Wilkinson, S., Emery, A.M., Khamis, I.S., Mgeni, A.F., Stothard, J.R., Rollinson, D., 2007. Spatial and temporal population genetic survey of *Bulinus globosus* from Zanzibar: an intermediate host of *Schistosoma haematobium*. J. Zool. 272, 329–339.
World Health Assembly, 2001. Fifty-fourth World Health Assembly, Geneva, 14–22 May 2001: Resolutions and Decisions. World Health Organization, Geneva.
World Health Organization, 1985. The Control of Schistosomiasis; Report of a WHO Expert Committee. Technical report series N°728. *From* Geneva 114.
World Health Organization, 2006. Communicable Disease Surveillance and Response Systems. Guide to Monitoring and Evaluation [Online]. World Health Organization, Geneva. Available: ⟨https://apps.who.int/iris/handle/10665/69331⟩ [Accessed].
World Health Organization, 2013. Schistosomiasis Progress Report 2001–2011 and Strategic Plan 2012–2020 [Online]. World Health Organization. Available: ⟨https://apps.who.int/iris/handle/10665/78074⟩ [Accessed].
World Health Organization, 2017. Field Use of Molluscicides in Schistosomiasis Control Programmes - An Operational Manual for Programme Managers. *From* 50.
World Health Organization, 2019. Guidelines for Laboratory and Field Testing of Molluscicides for Control of Schistosomiasis. World Health Organization, Geneva.
World Health Organization, 2020. Ending the Neglect to Attain the Sustainable Development Goals - A Road Map for Neglected Tropical Diseases 2021–2030 [Online]. World Health Organization. Available: ⟨https://apps.who.int/iris/handle/10665/338565⟩ [Accessed].
World Health Organization, 2022. WHO Guideline on Control and Elimination of Human Schistosomiasis [Online]. World Health Organization, Geneva. Available: ⟨https://www.who.int/publications/i/item/9789240041608⟩ [Accessed].
World Health Organization, 2023a. Schistosomiasis Key Facts [Online]. Available: ⟨https://www.who.int/news-room/fact-sheets/detail/schistosomiasis⟩ [Accessed 06.04.2023].
World Health Organization, 2023b. 34th meeting of the International Task Force for Disease Eradication, 19–20 September 2022. *Weekly Epidemiological Record*.
Xing, Y.-T., Dai, J.-R., Yang, K., Jiang, T., Jiang, C.-G., Mohammed, S.J., et al., 2021. *Bulinus* snails control by China-made niclosamide in Zanzibar: experiences and lessons. In: Yang, K., Mehlhorn, H. (Eds.), Sino-African Cooperation for Schistosomiasis Control in Zanzibar. Sprinter Nature, Switzerland.
Xue, C.G., Taylor, M.G., Bickle, Q.D., Saviol, L., Renganathan, E.A., 1993. Diagnosis of *Schistosoma haematobium* infection: evaluation of ELISA using keyhole limpet haemocyanin or soluble egg antigen in comparison with detection of eggs or haematuria. Trans. R. Soc. Trop. Med. Hyg. 87, 654–658.

Yang, K., 2021. Preface. In: Yang, K., Mehlhorn, H. (Eds.), Sino-African Cooperation for Schistosomiasis Control in Zanzibar. Springer, Cham, Switzerland.

Yang, K., Juma, S., Kabole, F., J, G., Garba, A., He, J., et al., 2021. China-Zanzibar cooperation project of schistosomiasis control: study design. In: Yang, K., Mehlhorn, H. (Eds.), Sino-African Cooperation for Schistosomiasis Control in Zanzibar. Springer, Cham, Switzerland.

Yang, K., Yang, H.T., Liang, Y.S., Dai, J.R., Li, W., Zhang, J.F., et al., 2019. A path analysis on China's participation in global health governance: a case study of china aid of schistosomiasis control in Zanzibar. Chin. J. Schistosomiasis Control 31, 14–18.

Zanzibar Protectorate, 1930–1934. Annual Report on the Medical and Sanitary Divisions for the Year 1929–1933. *From* Wellcome Collection.

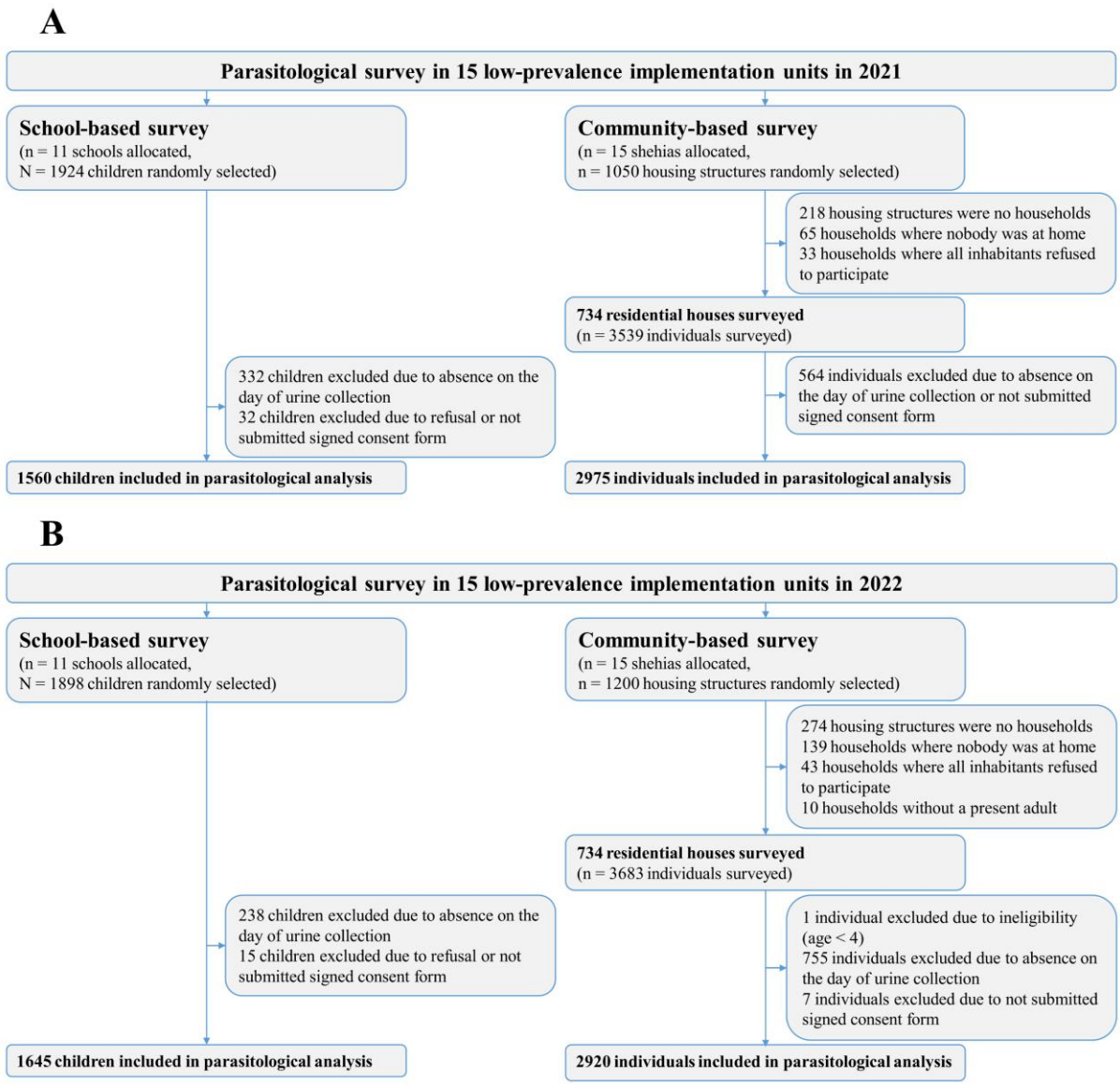
Zhang, S.-M., Bu, L., Lu, L., Babbitt, C., Adema, C.M., Loker, E.S., 2022. Comparative mitogenomics of freshwater snails of the genus *Bulinus*, obligatory vectors of *Schistosoma haematobium*, causative agent of human urogenital schistosomiasis. Sci. Rep. 12, 5357.

Zhou, L.Y., Luo, E.P., Yang, K., 2019. Current status of international discourse of china aid project of schistosomiasis control in Zanzibar. Chin. J. Schistosomiasis Control 31, 532–534.

Zhou, X.-N., Bergquist, R., Tanner, M., 2013. Elimination of tropical disease through surveillance and response. Infect. Dis. Poverty 2, 1.

Appendix B: Supplementary material from publication "Test-Treat-Track-Test-Treat (5T) approach for *Schistosoma haematobium* elimination on Pemba Island, Tanzania"

Supplementary File 1: Fig 1. Flow diagram of individuals participating in the school-based and household-based surveys in low *Schistosoma haematobium* prevalence shehias on Pemba, Tanzania, in 2021 (A) and 2022 (B).



Supplementary File 2: Table 1. Prevalence and intensity of microhematuria and *S. haematobium* of participants in school-based and household-based surveys 2021 and 2022, and test-treat-track-test-treat (5T) activities in 2021. NA = Not applicable

		2021 school-based survey		**2021 household-based survey**		**2021 school testing**		**2021 madrassa testing**		**2021 household tracking**		**2021 water body tracking**		**2021 health facilities**		**2022 school-based survey**		**2022 household-based survey**	
N		1560		2975		3700		594		258		60		354		1645		2920	
	female	839		1570		1942		284		152		27		201		855		1617	
	male	721		1405		1758		310		106		33		153		790		1303	
Age (median)		10		17		11		9		16.5		13		20		10		18	
Microhaematuria positive, n (%)		47	(3.1)	162	(5.5)	190	(5.1)	47	(8.0)	60	(23.3)	19	(31.7)	74	(20.9)	104	(6.3)	386	(13.2)
	female	27		110		105		27		32		11		36		55		249	
	male	20		52		85		20		28		8		38		49		137	
Age (median)		10		23		12		8		11		20		22		11		21	
Trace of microhaematuria, n (%)		26	(1.7)	64	(2.2)	50	(1.4)	13	(2.2)	16	(6.2)	9	(15.0)	4	(1.1)	80	(4.9)	227	(7.8)
Small microhaematuria (+), n (%)		5	(0.3)	28	(0.9)	33	(0.9)	10	(1.7)	13	(5.1)	4	(6.7)	5	(1.4)	7	(0.4)	77	(2.6)
Moderate microhaematuria (++), n (%)		10	(0.7)	39	(1.3)	47	(1.3)	12	(2.0)	9	(3.5)	3	(5.0)	18	(5.1)	8	(0.5)	49	(1.7)
Large microhaematuria (+++), n (%)		6	(0.4)	31	(1.0)	60	(1.6)	12	(2.0)	22	(8.6)	3	(5.0)	47	(13.3)	9	(0.5)	33	(1.1)
Urine filtration positive, n (%)		7	(0.5)	14	(0.5)	NA		NA		31	(12.8)	5	(8.5)	NA		6	(0.4)	19	(0.7)
	female	3		8		NA		NA		11		1		NA		2		9	
	male	4		6		NA		NA		20		4		NA		4		10	
Age (median)		9		10.5		NA		NA		8		18		NA		11		16	
Urine filtration heavy intensity, n (%)		2	(0.1)	1	(0.0)	NA		NA		10	(4.1)	1	(1.7)	NA		0	(0.0)	2	(0.1)