Distribution Agreement

In presenting this thesis or dissertation as a partial fulfillment of the requirements for an advanced degree from Emory University, I hereby grant to Emory University and its agents the non-exclusive license to archive, make accessible, and display my thesis or dissertation in whole or in part in all forms of media, now or hereafter known, including display on the world wide web. I understand that I may select some access restrictions as part of the online submission of this thesis or dissertation. I retain all ownership rights to the copyright of the thesis or dissertation. I also retain the right to use in future works (such as articles or books) all or part of this thesis or dissertation.

Signature:

_____ _____

Yuqing Wang Date

Associations between schistosome antibody status and incident HIV infection are not mediated by STIs / GUIs in a longitudinal cohort of Zambian men

By

Yuqing Wang

Master of Public Health

Infectious Disease Epidemiology Certificate

in Epidemiology

_____

Kristin M Wall, MS, PhD

Committee Chair

Associations between schistosome antibody status and incident HIV infection are not mediated by STIs / GUIs in a longitudinal cohort of Zambian men

By

Yuqing Wang

Bachelor of Science
Shanghai University of Traditional Chinese Medicine
2020

Thesis Committee Chair: Kristin M Wall, MS, PhD

An abstract of
A thesis submitted to the Faculty of the
Rollins School of Public Health of Emory University
in partial fulfillment of the requirements for the degree of
Master of Public Health
Infectious Disease Epidemiology Certificate
in Epidemiology
2023

Abstract

Associations between schistosome antibody status and incident HIV infection are not mediated by STIs / GUIs in a longitudinal cohort of Zambian men

By Yuqing Wang

Introduction. Schistosomiasis is an acute and chronic parasitic disease caused by parasitic worms. Schistosoma haematobium and Schistosoma mansoni are two main species causing human infections in Zambia. Some articles indicate that the prevalence of schistosomiasis in Zambia may be associated with increased HIV infection risk. However, causal mechanisms between schistosomiasis and HIV infections need to be understood. Sexually transmitted infections (STIs), and other causes of genital ulceration and inflammation (GUI) may mediate observed associations between schistosomiasis infection and HIV risk. In this study, we conducted an analysis to evaluate whether the effect of Schistosome-specific (SH) antibody status on incident HIV acquisition or transmission is mediated by STIs / GUIs among men, who were recruited in a longitudinal cohort of heterosexual HIV serodiscordant couples in Zambia.

Methods. We retrospectively analyzed data from men in a Zambian HIV-discordant couple cohort. Data and samples were collected in Lusaka, Zambia between 1994-2012 in a nested case-control design. SH antibody status was tested at baseline by enzyme-linked immunosorbent assay (ELISA). Descriptive analyses and associations between SH antibody status and baseline covariates were assessed, stratified by baseline HIV antibody status. Associations between SH antibody and time varying STIs outcomes were assessed, stratified by HIV antibody status. Multivariable logistic regression and survival analysis were applied.

Results. Of 1046 men, 65.97% (N=690) had a positive SH antibody status at baseline. Among 599 HIV+ men, SH antibody status at baseline was associated with higher viral load (crude odds ratio [cOR]=1.12, 95%CI [1.01,1.23], p=0.03), and SH antibody positivity of partners (cOR=1.80, 95%CI [1.21, 2.69], p=0.004). Among 447 HIV- men, SH antibody status at baseline was associated with SH antibody status of their partner (cOR=2.00, 95% CI [1.30, 3.08], P=0.002). No significant associations were detected between SH antibody and time varying STI/GUI outcomes.

Discussion. The data from our cohort do not support the hypothesis that STIs /GUIs are mediators between SH antibody status and HIV infection risk. Potential biases due to sub-optimal diagnostic tools for STI status, could affect the validity of our findings. Further studies are recommended to evaluate other mechanisms which explain the relationship among SH infections and HIV risk. This information could bolster the current strategy of HIV prevention in schistosomiasis endemic areas.

Associations between schistosome antibody status and incident HIV infection are not mediated by STIs / GUIs in a longitudinal cohort of Zambian men

By

Yuqing Wang

Bachelor of Science, Nursing
Shanghai University of Traditional Chinese Medicine
2020

Thesis Committee Chair: Kristin M Wall, MS, PhD

A thesis submitted to the Faculty of the
Rollins School of Public Health of Emory University
in partial fulfillment of the requirements for the degree of
Master of Public Health
Infectious Disease Epidemiology Certificate
in Epidemiology
2023

Table of Contents

Chapter I: Literature Review

Schistosomiasis Epidemiology

Schistosomiasis is an acute and chronic parasitic disease caused by parasitic worms, regarded as one of the most common neglected tropical diseases (NTDs) in the world. Infection usually occurs when contacting fresh water plagued by schistosomes, and schistosomes consequently penetrate the skin. Schistosomes will cause various gastrointestinal and urogenital symptoms which can contribute to anemia, malnutrition, and even death if the schistosome eggs cannot be passed out of bodies (Bartlett et al., 2022). Acute symptoms can manifest after an incubation period of 2-6 weeks after exposure. Commonly, young adolescents are the most vulnerable to schistosome infections. Schistosomiasis can also persist infecting adults who have high contact with contaminated water infested with schistosomes during specific activities, such as bathing, fishing, and laundry. Schistosomiasis has been becoming prevalent in tropical and subtropical areas especially in places where there is a shortage of clean water and adequate sanitations, and healthcare facilities. Schistosoma haematobium, and S. mansoni are two major schistosome species in Sub-Saharan Africa, which can manifest urogenital schistosomiasis and intestinal schistosomiasis respectively. In fact, Sub-Saharan Africa is suffering from a huge health burden of schistosomiasis, accounting for 90% of schistosome-infected cases with annually 280,000 deaths (van der Werf et al., 2003).

Urogenital schistosomiasis (UGS) is associated with the infection of Schistosome eggs. Male genital schistosomiasis (MGS) can result in a range of symptoms, including pelvic, coital, ejaculatory pain, abnormal ejaculates, haemospermia, abnormal swelling of genital organs, and

infertility (Kayuni et al., 2021). Although studies usually emphasized the adverse effects of schistosomiasis on female genital schistosomiasis (FGS) because of specific genital mucosal breach, increased abnormal vasculature, and elevated inflammatory cells in genital organs which may increase the risk of HIV acquisition and transmission (Kjetland et al., 2006), certain HIV-related mediators have also been detected in male genital organs that have similar risk of HIV infections compared with females' (Leutscher et al., 2005). Unfortunately, the influence of MGS in endemic areas is usually underestimated because of being misdiagnosed as other diseases, such as sexual transmitted illness (STI) (Kayuni et al., 2019). A study (Mbabazi et al., 2011) also indicated that chronic MGS will amplify the risk of HIV by increasing the HIV-1 viral load in semen, thereby elevating the risk of HIV transmissions to female sexual partners.

Schistosomiasis and HIV

Some evidence (Kleppa et al., 2014; Stecher et al., 2015) has shown that there is a high prevalence of coinfections of HIV and schistosomiasis in sub-Saharan Africa where over 24 million people are suffering from the geographic overlap of two diseases. UGS has been linked with higher risk of HIV infection in the previous study (Kayuni et al., 2021). For example, FGS can amplify the risk of HIV infections among women resulting from chronic inflammatory lesions, genital epithelial bleeding and sandy patches in the cervix and vagina caused by schistosome eggs in genital organs (Aula et al., 2021). For males, a potential mechanism has been hypothesized for MGS that mechanical breach of the genital mucosa and local recruitment of CD4+ cells in male genitals make it more susceptible to HIV infections and transmissions during intercourse (Stecher et al., 2015). Also, increased genital vascularity due to MGS, is linked with higher leukocyte counts in semen, hence increasing seminal HIV viral load resulting in higher HIV transmissions (Kjetland

et al., 2014). However, inadequate quantity and quality of studies can support the specific mechanisms between MGS and HIV in SH-endemic regions. While most studies have concentrated on studying FGS cases solely, MGS is still neglected. One main reason for this dilemma is that accurate diagnostic criteria of MGS have not been established because of few available diagnostic tools in poor communities. In fact, a longitudinal study found that women were more prone to HIV virus from HIV+ male partners among HIV serodiscordant couples in Zambia (Wall et al., 2018). As a result, Specific information including immunological background and mucosal changes should also be analyzed on MGS to promote HIV preventive strategy for heterosexual couples (Stecher et al., 2015).

Mechanisms

Many studies indicated the association between SH infection and HIV, and it was considered that UGS caused by SH infected status increased the risk of HIV transmission and acquisition (Kleppa et al., 2014; Mbabazi et al., 2011; Wall et al., 2018). The casual pathway between SH infection and HIV remains uncertain. Genital ulceration and inflammation (GUI) and sexual transmitted infections (STIs) have been regarded as important transmission co-factors of HIV transmission since 1990s (Cameron et al., 1989; Laga et al., 1993; Plummer et al., 1991).

Because of similar symptoms arise from STI/GUIs and UGS, such as Genital discharges or inflammation, elevated leukocyte cell counts in genitals. Some studies postulated that STIs/GUIs may mediate the effects of SH infections on HIV (Downs et al., 2017; Wall et al., 2018) (Fig 1). Previous data has found that among women, the increase of cervicovaginal proinflammatory cytokines associated with STIs or bacterial vaginosis (BV), is related with higher viral load set point,

which is predictive of time to AIDS (Roberts et al., 2012). Furthermore, SH -positive women with primary or secondary syphilis, or herpes simplex virus (HSV) type-1 pr -2 infections, had higher number of activated T-cells in the genital tract, which increased the susceptibility of HIV infections (Rebbapragada et al., 2007). In addition to men, it has been demonstrated that MGS is an important risk factor for HIV transmission since it can increase the leukocyte count and density of HIV target cells including CD4+ T-lymphocytes (Leutscher et al., 2005), However, few studies have investigated the effect of SH infection on HIV can be mediated by STI/GUIs neither among women nor men.

Prevention and control

According to WHO guidelines, the schistosome preventive strategy in SH-endemic regions mainly consists of two aspects: large-scale treatments of risk groups, and management of contacts and environment. In terms of treatments, praziquantel (PZQ) is recommended for treating all forms of schistosomiasis since it is a safe, inexpensive, and nontoxic medication that is suitable for not only pregnant women and young children, and it has been applied for treatment and control of schistosomiasis over 40 years (Caffrey, 2015; Mbabazi et al., 2011). In this case, regular drug treatments with PZQ should be conducted for susceptible populations, including pre-school-aged / school-aged children, adults with occupational exposures of water infested with schistosome, and any others in SH-endemic areas. Despite the potential resistance of PZQ and the racemic nature of PZQ meaning that massive doses are required to be pharmacologically useful (Caffrey, 2015), PZQ is still a reliable treatment of schistosomiasis. In terms of management of contacts and environment, schistosome re-infection is still threatening people's health in SH-endemic areas due to poor/no adherence to PZQ treatments and low

coverage, contaminated water sources due to climatic conditions, and high exposure of contaminated water because of occupational requirements or daily activities (Aula et al., 2021). Hence, an integrated intervention of management of contacts and environment is required, including: snail control; persistent PZQ treatment; active risk mapping and disease surveillance; accurate diagnosis of schistosomiasis; promoting access to clean water, and sanitations; SH-related educations about changing risky behaviors in order to prevent infections (Cioli et al., 2014). Overall, a multifaceted approach of schistosome treatment and prevention is essential to help achieve the goal of schistosome eliminations in Africa.

Technical and socioeconomic limitations

The paucity of epidemiological studies about schistosomiasis in Africa has hindered the process of schistosomiasis control and elimination. Technical and socioeconomic factors contribute to this dilemma.

In the context of technical factors, the diagnosis of MGS remains underestimated and even misunderstood due to lack of accurate diagnostic techniques and approaches (Kayuni et al., 2019). According to previous studies, several diagnostic techniques have been recommended for accurate MGS diagnosis including: conducting microscopy on semen and biopsy material from genital lesions (Leutscher et al., 2008); eosinophil cationic protein (ECP), circulating anodic antigen (CAA), and soluble egg antigen (SEA) as blood-based markers of MGS; PCR and DNA-based tests (Leutscher et al., 2000; Leutscher et al., 2008); and ultrasonography in diagnosing genital lesions (Ramarakoto et al., 2008). However, all of them are too expensive to be applied in SH-endemic areas in Africa. As a result, it is difficult to establish standardized protocols for

analysis of semen. Further research is required to explore the availability of low-cost techniques to diagnose MGS.

Low socioeconomic conditions also exacerbate the harm of SH infections. For instance, sub-optimal treatment compliance due to social stigmatization of schistosomiasis (Lothe et al., 2018), or wrongly regarding SH infections as normal signs of puberty (Boko et al., 2016) reduced the effectiveness of PZQ treatment, and contributed to the high prevalence of SH infections especially among adolescents. In addition, gender disparities in African society blurred the true health burden of schistosomiasis. African women generally have lower economic and social status than men, and the disease occurrence of schistosomiasis in females is usually underreported (Wharton-Smith et al., 2019). Furthermore, insufficient healthcare infrastructure due to poor economic conditions, weaken the efficacy of disease surveillance so that effective control interventions are hard to be conducted (Aula et al., 2021).

Summary

A better understanding of the relationship between SH infections and HIV, and potential mediators of this relationship, is crucial for improved HIV prevention (Fig 1). More broadly, a better understanding of MGS treatment and prevention is needed, as well as how to integrate MGS and HIV services in Africa.

Chapter II: Manuscript

Associations between schistosome antibody status and incident HIV infection are not mediated by STIs / GUIs in a longitudinal cohort of Zambian men

By Yuqing Wang

Introduction. Schistosomiasis is an acute and chronic parasitic disease caused by parasitic worms. Schistosoma haematobium and Schistosoma mansoni are two main species causing human infections in Zambia. Some articles indicate that the prevalence of schistosomiasis in Zambia may be associated with increased HIV infection risk. However, causal mechanisms between schistosomiasis and HIV infections need to be understood. Sexually transmitted infections (STIs), and other causes of genital ulceration and inflammation (GUI) may mediate observed associations between schistosomiasis infection and HIV risk. In this study, we conducted an analysis to evaluate whether the effect of Schistosome-specific (SH) antibody status on incident HIV acquisition or transmission is mediated by STIs / GUIs among men, who were recruited in a longitudinal cohort of heterosexual HIV serodiscordant couples in Zambia.

Methods. We retrospectively analyzed data from men in a Zambian HIV-discordant couple cohort. Data and samples were collected in Lusaka, Zambia between 1994-2012 in a nested case-control design. SH antibody status was tested at baseline by enzyme-linked immunosorbent assay (ELISA). Descriptive analyses and associations between SH antibody status and baseline covariates were assessed, stratified by baseline HIV antibody status. Associations between SH antibody and time

varying STIs outcomes were assessed, stratified by HIV antibody status. Multivariable logistic regression and survival analysis were applied.

Results. Of 1046 men, 65.97% (N=690) had a positive SH antibody status at baseline. Among 599 HIV+ men, SH antibody status at baseline was associated with higher viral load (crude odds ratio [cOR]=1.12, 95%CI [1.01,1.23], p=0.03), and SH antibody positivity of partners (cOR=1.80, 95%CI [1.21, 2.69], p=0.004). Among 447 HIV- men, SH antibody status at baseline was associated with SH antibody status of their partner (cOR=2.00, 95% CI [1.30, 3.08], P=0.002). No significant associations were detected between SH antibody and time varying STI/GUI outcomes.

Discussion. The data from our cohort do not support the hypothesis that STIs /GUIs are mediators between SH antibody status and HIV infection risk. Potential biases due to sub-optimal diagnostic tools for STI status, could affect the validity of our findings. Further studies are recommended to evaluate other mechanisms which explain the relationship among SH infections and HIV risk. This information could bolster the current strategy of HIV prevention in schistosomiasis endemic areas.

Introduction

Schistosomiasis is a disease caused by parasitic worms, regarded as one of the most common neglected tropical diseases (NTDs) in the world. Schistosoma mansoni, Schistosoma haematobium, and Schistosoma Japonicum cause the majority of human infections. According to the CDC, schistosomiasis has infected more than 200 million people all over the world.

Schistosomiasis can be transmitted to humans via contaminated freshwater and cercariae (free-swimming larval forms) via snails and can penetrate human skin and develop into adult worms. School-age children are usually at risk when they are swimming or bathing in contaminated water having cercariae. The adult worms can be lodged in the intestine or bladder and cause inflammation or scaring if they are not able to pass out of the body. Symptoms are usually associated with human immune responses to the worms and schistosome eggs. According to the CDC, fever, chills, cough, and muscle aches are common symptoms; chronic infections can be attributed to more severe symptoms, such as liver fibrosis or bladder cancer. Some people may be asymptomatic after infected. However, schistosome worms and eggs can lodge in human tissues for long periods of time, and individuals can excrete eggs potentially leading to new infections.

Africa has been suffering from huge health burdens of schistosomiasis, accounting for 70 million disability-adjusted life years (DALYs) lost and approximately 300,000 deaths annually (King & Dangerfield-Cha, 2008; van der Werf et al., 2003). Some evidence suggests that high risks of schistosomiasis may be related to increasing temperatures, especially in tropical areas, although the specific effects of climate change on schistosomiasis infections remains uncertain

(Yang et al., 2007). In addition to climate aspects, factors such as urbanization trends seem to be alter the risk of schistosome infections (van der Werf et al., 2003).

Schistosome infections may be associated with HIV transmission risk and severe HIV outcomes. A study in Zambia indicated that prevalence of schistosome infections was related to increased HIV transmission risk in both sexes and HIV acquisitions and earlier death among HIV+ women (Wall et al., 2018). Another study of Tanzanian women also found that the odds of HIV infection among women with S. haematobium infections was four times higher than for women without S. haematobium (Downs et al., 2011). Women may be more vulnerable to HIV-1 infections compared to men because schistosome eggs affect the mucosal tissue of the vagina and cervix which increases the risk of HIV infection during sexual exposures, while schistosome eggs rarely impact male external genital organs (Cheever et al., 1977; Edington et al., 1975; Jourdan et al., 2011).

Male genital schistosomiasis (MGS) is a specific manifestation of urogenital schistosomiasis (UGS), correlated with genital inflammations or fibrosis due to the presence of schistosome eggs in the genitalia of men. Pelvic, coital or ejaculatory pain, abnormal ejaculates, haemospermia, abnormal swelling of genital organs and infertility are common consequences of MGS (Kayuni et al., 2021). Previous data have connected STIs and genital ulceration and inflammation (GUI) with HIV acquisition (Wall et al., 2017). However, a review (Kayuni et al., 2019) specified that hypothesized associations between MGS and STIs as well as between MGS and HIV transmissions remain unstudied. Whether schistosome infection in men affects the risk of HIV acquisition or transmission mediated by STIs / GUIs is currently unknown.

It is essential to understand the causal pathway between schistosome infections, STIs/GUIs, and HIV infections. According to World Health Organization (WHO) recommendations, praziquantel is suggested for treating most schistosome infections since it is effective, cheap, and safe. Increasing evidence indicates the plausible association between urogenital schistosomiasis and HIV infections. Integrating anti-Schistosoma treatments and HIV prevention treatments may have significant effects on millions of people in Africa where schistosomiasis and HIV are co-endemic (Mbabazi et al., 2011). In addition, if STIs and GUIs mediate this causal pathway, corresponding STI/GUI prevention strategies should also be applied to promote the effectiveness of integrated treatments.

In our retrospective analysis of a heterosexual HIV-discordant couple prospective cohort in Lusaka, the capital of Zambia, conducted from 1994 to 2009, we hypothesized that the association between schistosome infections and HIV acquisition or transmission is mediated by STIs / GUIs among men. S. mansoni, S. haematobium are two major schistosome species in Zambia, a country in southern Africa.

Methods

Ethics Statement

The cohort study was approved by the Office for Human Research Protections-registered Institutional Review Boards of Emory University and University of Zambia. All participants were recruited after providing informed consent. All participants were deidentified during data collection and engaged investigators had no access to or direct contact with participants.

Study Participants

The cohort study included heterosexual HIV discordant couples (M+F-, M-F+) in Lusaka, Zambia enrolled between 1994 – 2009. Serodiscordant couples identified via couple's voluntary counselling and testing (CVCT) services in Lusaka, Zambia were enrolled in quarterly follow-up.(Wall et al., 2012) CVCT provided group counselling, joint counselling including confidential informed consent procedures, phlebotomy, rapid HIV testing, and joint post-test counselling and test result delivery (Boeras et al., 2011). Eligible couples received consistent services from CVCT during the follow up period. Couples were censored given antiretroviral treatment initiation, if either partner died, or if the relationship dissolved. We retrospectively sampled all eligible participants for SH antibody testing of stored plasma samples in a nested case-control design.

Outcome of interests: HIV acquisition or transmission

HIV- partners of all discordant couples at enrollment received HIV rapid antibody tests every 1-3 months, and the corresponding HIV incidence was measured consistently (Boeras et al., 2011). We defined the date of infection as the minimum of the midpoint between the last negative and first positive antibody test; either two weeks before a first antigen positive test or a first viral load antibody test (Wall et al., 2018).

Mediators of interests: time varying sexually transmitted infections (STIs) and other genital abnormalities

Our time-varying mediators included non-STI genital inflammation (defined as clinical or laboratory diagnosis or treatment of gonorrhea, chlamydia, or trichomonas), STI genital inflammation (defined as reported discharge, dysuria, dyspareunia; observed discharge or

inflammation of external or internal genitalia) (Wall et al., 2017), bilateral inguinal adenopathy, and genital ulcer. Mediators were measured at baseline and every follow-up visit.

Exposure of interests: Schistosome-specific antibody status

The cohort study collected blood plasma and serum samples from all couples at enrollment and stored them in a repository at Emory University. In 2010, samples were retrospectively tested from individuals enrolled between 1994 and 2009 for antibodies by schistosome soluble worm antigen preparation (SWAP) using enzyme-linked immunosorbent assay (ELISA) (Shane et al., 2011). ELISA results were not available for every couple. All seroconverted individuals along with non-seroconverted individuals were sampled from the original cohort and included in this nested case-control study. To ensure consistency between plates for antibody testing, a 1:3 serial dilution curve was made, a four-parameter curve fitting model was used, and the positive cutoff value was set at two standard deviations above the average anti-SWAP IgG value of 13 different samples (Shane et al., 2011).

Demographic and baseline covariates

Descriptive data of individuals included demographics (age, years of cohabiting, monthly household income (USD), Fertility intentions) clinical covariates (HIV stage if HIV+, viral load if HIV+, herpes simplex virus type 2 (HSV-2), having any past-year history of any STIs, RPR status (any syphilis infections), and SH antibody status of partners). Baseline data were grouped by SH antibody status stratified by HIV acquisition status.

Data analysis

Analyses were conducted by SAS v9.4. Descriptive analyses were conducted for baseline data. Categorical covariates were summarized by counts and percentages, and continuous covariates were described by either means and standard deviations, or medians with the lower and upper quartiles depending on the distributions of covariates. Statistical analyses were performed for baseline data: we conducted univariable logistic regression to estimate the odds ratios and corresponding 95% confidence intervals between SH antibody status and baseline covariates stratified by HIV acquisition status; chi-square and t-tests were used for testing the statistical significance of baseline covariates. Univariable proportional hazard (PH) cox models were conducted for estimating crude hazard ratios and corresponding 95% confidence intervals between time varying outcomes and SH antibody status; we validated that SH antibody status met PH assumptions derived from results of log-log plots, plots based on martingale residuals, and goodness of fit tests.

Results

We analyzed 1046 men from HIV discordant couples in this nested case-control study. 63.60% of HIV+ males (N=381) had positive SH antibody status at baseline, compared to 69.13% of HIV- males (N=309) had positive SH antibody status at baseline. No significant differences were detected in age, years of cohabiting, monthly household income (USD), Fertility intentions in demographic and family planning characteristic. Among clinical characteristics, significant associations with SH antibody status were detected in VL (cOR=1.12, 95% CI [1.01,1.23], p=0.03) among HIV+ males; and SH antibody status of partner was significantly associated with SH

antibody status among HIV+ males, (cOR=1.80, 95% [1.21,2.69], p=0.004), and among HIV- males, (cOR=2.00, 95%[1.30,3.08], P=0.002) (Table 1a, Table 1b).

However, no significant associations were detected among time varying STIs / GUIs and SH antibody status (Table 2a, Table 2b).

Discussion

Among all HIV+ men in this cohort, participants with positive SH antibody at baseline have significantly higher viral loads than men with negative SH antibody at baseline (Table 1a). Furthermore, the odds of HIV transmission to HIV- partners among men infected by schistosomiasis is significantly higher than that among men not infected by schistosomiasis (Table 1b). These findings support the idea in the previous publication (Leutscher et al., 2005) that among men co-infected by schistosomiasis and HIV, schistosome egg excretion in semen may be related with increasing cells contributing to HIV replication. In this scenario, preventing schistosomiasis could be attributed to controlling HIV epidemic in schistosomiasis endemic regions.

Although we hypothesized that time varying STIs / GUIs can be potential mediators between SH antibody status and HIV incidence and acquisition observed in previous studies (Downs et al., 2017; Wall et al., 2018) (Fig 1), we did not find that STIs / GUIs were statistically significant or meaningful mediators. Among 1046 men in this cohort with an average follow-up time of two years, time-varying outcomes, including non-STI genital inflammation, STI genital inflammation, bilateral inguinal adenopathy, and genital ulcer, were not associated with SH antibody status at baseline regardless of HIV infection status. Schistosome eggs rarely impact

male external genital organs based on the previous study (Cheever et al., 1977), which may explain why no direct association between SH antibody status and STIs / GUIs was detected in our study. However, we observed a general trend that men infected by schistosomiasis had more time varying STI / GUIs outcomes than men without schistosome infection although the results were not statistically significant ($p > 0.05$). As a result, it is reasonable to presume that schistosome infection could increase the risk of STI / GUIs.

A few mechanisms have been reported in the previous publications (Leutscher et al., 2005; Stecher et al., 2015) that the mechanical breach of genital mucosa and higher genital vascularity caused by MGS will render individuals to local recruitment of CD4+ cells and higher leukocyte frequencies in semen, which results in higher risks of HIV acquisition and transmission during intercourse. ELISA tests measured SH antibody status at baseline, which is not the gold standard for diagnosing current MGS, but rather reflects past or current infection. Urine microscopy, PCR, and DNA-based tests can be reliable ways to diagnose current MGS (Le and Hsieh, 2017). Also, ultrasonography can be useful for diagnosing lesions due to MGS (Ramarakoto et al., 2008), but the corresponding machines of ultrasonography are still not available enough in endemic areas because of high prices. Due to these conditions, MGS is still underreported, and is usually mistaken by STIs (Kayuni et al., 2019). The results of our study might be biased due to misclassification of SH or STI/GUI infection. As a result, distinguishing MGS from STIs/GUIs among men is essential to help identify the modified effects of potential mediators between schistosomiasis and HIV.

There are some limitations in our study. Firstly, we did not apply gold standard tests as recommended (Shane et al., 2011) for detecting active schistosome infections, but ELISA

antibody testing retrospectively for a subset of the total cohort to detect both current and past infections. Also, we were not able to distinguish time varying outcomes whether caused by MGS or STIs / GUIs due to limited diagnostic techniques. In addition, the number of time varying STI/GUIs were limited which reduces the power of our statistical analysis. In the further studies, more accurate diagnosis of current MGS infection via microscopy, DNA-based tests, or biopsy on genital lesions is recommended to reduce potential misclassification biases of SH antibody status due to MGS which could be misclassified as STIs / GUIs. Also, a larger sample size is preferred to improve the power.

Conclusions

The mechanism underlying the association between MGS and HIV risk is unclear, and an improved understanding of this mechanism could inform improved HIV prevention strategies.

Chapter III: Public Health Recommendations

Schistosomiasis is a neglected tropical disease (NTD) affecting over 240 million people around the world, 90% of whom are in Africa (Patel et al., 2021). SH infections are prevalent in low-income developing countries with limited sources of clean water, sanitation and hygiene techniques, and healthcare facilities. The integrated components of SH prevention and control should include: the large-scale of anthelmintic therapy of populations at risk; developing adequate sanitation and hygiene techniques; and increasing access to clean water (Bartlett et al., 2022). People are still suffering from huge burdens of schistosomiasis in Africa regardless of great efforts made on SH prevention and control by local governments.

Male genital schistosomiasis (MGS) is a specific manifestation of urogenital schistosomiasis (UGS) resulting from the presence of Schistosome eggs and related pathologies in genitalia of men (Leutscher et al., 2000). Previous publication suggests that individuals diagnosed UGS are susceptible to HIV infections and easier to transmit HIV to others (Mbabazi et al., 2011). Sub-Saharan Africa (SSA) has been suffering from tremendous burden of HIV epidemic, accounting for over 70% of the global burden of it (Kharsany & Karim, 2016). 8.1 million (32%) of people living in the HIV still haven't been treated in SSA in 2021 even though with great progress of antiretroviral therapy achieved and billions of dollars budgeted for HIV prevention and control among countries in SSA (Gupta et al., 2022; Kharsany & Karim, 2016). With a salient overlap of SH and HIV infections in SSA, intervening SH endemic may improve the effectiveness of HIV interventions and somehow ameliorate the impairments induced by HIV endemic in SSA. However, standardized approaches of diagnostic techniques and case managements for MGS

haven't been established yet due to limited research, socioeconomic limitations, and neglectful attitudes of MGS in the past decades (Kayuni et al., 2021).

Demonstrating the specific mechanism between MGS and HIV can strongly impact not only health of males but of females in SSA. Some study pointed out that MGS might not increase the HIV acquisition through local effects, such as genital lesions, but rather through schistosomiasis-related immunomodulatory effects, such as higher HIV viral loads concentrating in seminal fluids (Leutscher et al., 2005). It might partly explain why we could not detect the significant correlation of MGS and sexually transmitted illness (STI), or Genital ulceration and inflammation (GUI). However, males can still be potential carriers of HIV, thereby increasing the risk of HIV acquisition of their sexual partners. On the other hand, among women, SH infections may increase the risk of HIV acquisitions and transmissions not only because of local genital effects but chronic immunomodulatory effects (Mbabazi et al., 2011), and SH infections among adolescent girls and young women primary contribute to endemic schistosomiasis in SSA (Patel et al., 2021). In this case, it is plausible to postulate that intervening MGS endemics play an essential factor of controlling HIV new incidences among both males and females in SSA.

In the prospective of public health, budgeting the diagnostic techniques of MGS may be one of the important aspects to intervene MGS endemics. As suspicious lesions/tissues usually exist in semen, microscopy, PCR and DNA based tests would be preferred for accurate diagnosis of MGS. Expensive costs make it difficult to standardize the gold standard of diagnosis, thus underreporting and underestimating the intensity of MGS endemics. Furthermore, improving levels of education in SH endemic areas may have positive impacts on SH and HIV prophylaxis. This may help solve the underreported estimates of schistosomiasis burdens especially among

females induced by gender disparity, illiteracy, unequal social status, stigmatization and misunderstanding of disease due to local culture beliefs (Amazigo et al., 1997; Wharton-Smith et al., 2019). In addition, a larger scale of praziquantel (PZQ) treatments in SH-endemic areas is also recommended. Previous research suggested that higher doses of PZQ could be more effective of MGS than traditional doses could (Lang et al., 2017), while receiving adequate doses of PZQ is usually unavailable for both adolescents and adults in SSA (Kayuni et al., 2019). Furthermore, risk mapping combined with environmental monitoring is recommended to help control repetitive outbreaks or infections (Aula et al., 2021).

Further study should detect the relationship of MGS and HIV based on gold-standard diagnostic approach of MGS. Meanwhile, the optimal dose of PZQ treatment should also be demonstrated to optimize the mass drug administrations in SH-endemic regions. Furthermore, interventions in terms educational improvement are necessary to be integrated in therapeutic interventions to improve adherence.

Tables and Figures

Figure 1. Diagram of relationship between baseline schistosome-specific (SS) antibody and HIV acquisition or transmission with potential confounders and effective modifiers.

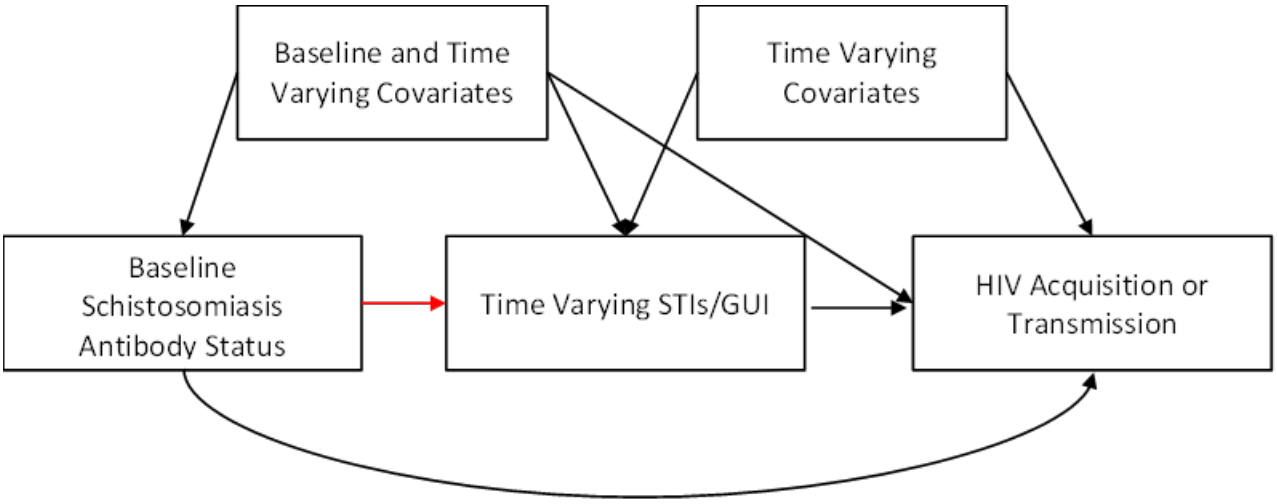

Table 1a. Descriptive analyses and associations between baseline covariates by SH antibody status among Zambian HIV+ men.

Table 1a. Descriptive analyses and associations between baseline covariates by SH antibody status among HIV+ Zambian men								
	HIV+ Men (N=599)							
	SH antibody positive (N=381)		**SH antibody negative**		**cOR**	**95%CI**		**p-value**
	N	**%**	**N**	**%**				
Demographics								
Age*	381	34.26 ± 7.59	218	34.22 ± 7.90	1.00	0.92	1.09	0.95
Years cohabiting***	375	6.00 (2.0,11.0)	214	5.00 (3.0,10.0)	1.03	0.95	1.13	0.46
Monthly household income (USD)***	375	42.00 (18.4, 69.6)	213	50.4 (28.8,84.0)	0.94	0.86	1.02	0.11
Family planning characteristics								
Fertility intentions	78	-	84	-	-	-	-	0.19
Yes, next year	19	24.36%	12	14.29%	1.71	0.73	4.00	-
Yes, but not next year	22	28.21%	32	38.10%	0.74	0.37	1.50	-
Don't know/No (ref)	37	47.44%	40	47.62%	-	-	-	-
Clinical characteristics								
HIV stage, if positive	381	-	218	-	-	-	-	0.79
Stage I (ref)	82	21.52%	50	22.94%	-	-	-	-
Stage II	145	38.06%	77	35.32%	1.15	0.73	1.80	-
Stage III-IV	154	40.42%	91	41.74%	1.03	0.67	1.60	-
VL, if positive (log10 copies/ml)*	229	4.98 ± 0.80	188	4.79 ± 0.87	1.12	1.01	1.23	0.03
HSV-2 status	284	-	123	-	-	-	-	0.49
Positive	206	72.54%	92	74.80%	1.12	0.60	2.08	-
Indeterminate	42	14.79%	13	10.57%	1.62	0.70	3.75	-
Negative (ref)	36	12.68%	18	14.63%	-	-	-	-
Past year history of any STI	380	-	217	-	-	-	-	0.31
Yes	178	46.84%	111	51.15%	0.84	0.60	1.18	-
No (ref)	202	53.16%	106	48.85%	-	-	-	-
RPR status of man	284	-	142	-	-	-	-	1.00
Positive	4	1.41%	2	1.41%	1.00	0.18	5.53	-
Negative (ref)	280	98.59%	140	98.59%	-	-	-	-
SH antibody status of partner	329	-	139	-	-	-	-	0.004
Positive	197	59.88%	63	45.32%	1.80	1.21	2.69	-
Negative (ref)	132	40.12%	76	54.68%	-	-	-	-

Index:

SH: Schistosome-specific; USD: United States Dollar; STI: sexually transmitted infection; OR: odds ratio; CI: confidence interval; HSV-2: herpes simplex virus 2; VL: viral load; RPR: rapid plasma regain.

viral loads collected from 1999

p-values are two-tailed

* Indicates continuous variable, mean and standard deviation reported

**Circumcised at baseline or ever during follow-up

***Median and interquartile range reported

Table 1b. Descriptive analyses and associations between baseline covariates by SH antibody status among Zambian HIV- men.

Table 1b. Descriptive analyses and associations between baseline covariates by SH antibody status among HIV- Zambian men								
	HIV- Men (N=447)							
	SH antibody positive (N=309)		**SH antibody negative (N=138)**		**cOR**	**95%CI**		**p-value**
	N	**%**	**N**	**%**				
Demographics								
Age*	308	35.13 ± 9.19	138	33.47 ± 8.66	1.02	0.99	1.05	0.07
Years cohabiting***	304	3.00 (1.0,7.0)	138	4.00 (2.0, 7.0)	1.01	0.98	1.04	0.64
Monthly household income (USD)***	307	42.00 (21.0,76.0)	138	49.43 (26.10, 77.52)	0.999	0.997	1.001	0.36
Family planning characteristics								
Fertility intentions	65	-	35	-	-	-	-	0.43
Yes, next year	10	44.62%	6	17.14%	0.63	0.19	2.16	-
Yes, but not next year	26	40.00%	18	51.43%	0.55	0.22	1.37	-
Don't know/No (ref)	29	15.38%	11	31.43%	-	-	-	-
Clinical characteristics								
HIV stage, if positive	-	-	-	-	-	-	-	-
Stage I (ref)	-	-	-	-	-	-	-	-
Stage II	-	-	-	-	-	-	-	-
Stage III-IV	-	-	-	-	-	-	-	-
VL, if positive (log10 copies/ml)*	-	-	-	-	-	-	-	-
HSV-2 status	229	-	87	-	-	-	-	0.78
Positive	120	52.40%	43	49.43%	1.21	0.69	2.11	-
Indeterminate	42	18.34%	15	17.24%	1.21	0.58	2.52	-
Negative (ref)	67	29.26%	29	33.33%	-	-	-	-
Past year history of any STI	309	-	138	-	-	-	-	0.44
Yes	107	34.63%	53	38.41%	0.85	0.56	1.29	-
No (ref)	202	65.37%	85	61.59%	-	-	-	-
RPR status of man	269	-	119	-	-	-	-	0.81
Positive	3	1.12%	1	0.84%	1.33	0.14	12.93	-
Negative (ref)	266	98.88%	118	99.16%	-	-	-	-
SH antibody status of partner	285	-	122	-	-	-	-	0.002
Positive	188	66.96%	60	49.18%	2.00	1.30	3.08	-
Negative (ref)	97	34.04%	62	50.82%	-	-	-	-

Index:

SH: Schistosome-specific; USD: United States Dollar; STI: sexually transmitted infection; OR: odds ratio; CI: confidence interval; HSV-2: herpes simplex virus 2; VL: viral load; RPR: rapid plasma regain.

viral loads collected from 1999

p-values are two-tailed

* Indicates continuous variable, mean and standard deviation reported

**Circumcised at baseline or ever during follow-up

***Median and interquartile range reported

Table 2a: Association between SH antibody and time-varying outcomes among HIV+ men

Table 2a. Association between SH antibody status and time-varying outcomes among HIV+ Zambian men								
	HIV+ Men (N=599)							
	SH antibody positive (N=381)		**SH antibody negative (N=218) reference group**		**cHR**	**95%CI**		**p-value**
	N	**%**	**N**	**%**				
Time-varying outcomes								
Non-STI genital inflammation								0.68
Yes	42	1.41%	35	1.42%	0.87	0.47	1.64	-
No	2930	98.59%	2438	98.58%	-	-	-	-
STI genital inflammation								0.06
Yes	90	3.03%	40	1.62%	1.68	0.98	2.88	-
No	2882	96.97%	2433	98.38%	-	-	-	-
Bilateral inguinal adenopathy								0.96
Yes	340	15.73%	195	15.78%	1.01	0.78	1.31	-
No	1822	84.27%	1041	84.22%	-	-	-	-
Genital ulcer								0.84
Yes	565	18.99%	435	17.58%	0.98	0.77	1.23	-
No	2411	81.01%	2039	82.42%	-	-	-	-
Other time-varying confounders								
Sex with study partner without a condom any time during follow-up								0.50
Yes	1139	36.09%	925	32.87%	0.95	0.80	1.12	-
No	2017	63.91%	1889	67.13%	-	-	-	-
More than one sex partner ever during follow-up								0.06
Yes	373	11.79%	499	17.44%	0.59	0.34	1.03	-
No	5154	88.21%	2362	82.56%	-	-	-	-

SH: schistosome-specific; STI: sexually transmitted infection; cHR: crude hazard ratio.

CI: confidence interval

Table 2b: Association between SH antibody and time-varying outcomes among HIV- men

Table 2b. Association between SH antibody status and time-varying outcomes among HIV- Zambian men								
	HIV- Men (N=447)							
	SH antibody positive (N=309)		**SH antibody negative (N=138) reference group**		**cHR**	**95%CI**		**p-value**
	N	**%**	**N**	**%**				
Time-varying outcomes								
Non-STI genital inflammation								0.80
Yes	83	3.68%	41	3.33%	1.07	0.65	1.76	-
No	2175	96.32%	1189	96.67%	-	-	-	-
STI genital inflammation								0.37
Yes	49	2.17%	19	1.54%	1.42	0.66	3.04	-
No	2209	97.83%	1211	98.46%	-	-	-	-
Bilateral inguinal adenopathy								0.43
Yes	253	14.90%	108	12.80%	1.15	0.82	1.61	-
No	1445	85.10%	736	87.20%	-	-	-	-
Genital ulcer								0.16
Yes	190	8.41%	94	7.64%	1.74	0.81	3.72	-
No	2070	91.59%	1136	92.36%	-	-	-	-
Other time-varying confounders								
Sex with study partner without a condom any time during follow-up								0.0503
Yes	948	41.95%	605	49.07%	0.85	0.73	1.00	-
No	1312	58.05%	628	50.93%	-	-	-	-
More than one sex partner ever during follow-up								0.64
Yes	311	13.71%	193	15.55%	0.88	0.52	1.49	-
No	1957	86.29%	1048	84.45%	-	-	-	-

SH: schistosome-specific; STI: sexually transmitted infection; cHR: crude hazard ratio.

CI: confidence interval

Reference

Amazigo, U. O., Anago-Amanze, C. I., & Okeibunor, J. C. (1997). URINARY SCHISTOSOMIASIS AMONG SCHOOL CHILDREN IN NIGERIA: CONSEQUENCES OF INDIGENOUS BELIEFS AND WATER CONTACT ACTIVITIES. *Journal of Biosocial Science*, *29*(1), 9-18. https://doi.org/10.1017/S0021932097000096

Aula, O. P., McManus, D. P., Jones, M. K., & Gordon, C. A. (2021). Schistosomiasis with a Focus on Africa. *Trop Med Infect Dis*, *6*(3). https://doi.org/10.3390/tropicalmed6030109

Bartlett, A. W., Sousa-Figueiredo, J. C., van Goor, R. C., Monaghan, P., Lancaster, W., Mugizi, R., Mendes, E. P., Nery, S. V., & Lopes, S. (2022). Burden and factors associated with schistosomiasis and soil-transmitted helminth infections among school-age children in Huambo, Uige and Zaire provinces, Angola. *Infect Dis Poverty*, *11*(1), 73. https://doi.org/10.1186/s40249-022-00975-z

Boeras, D. I., Luisi, N., Karita, E., McKinney, S., Sharkey, T., Keeling, M., Chomba, E., Kraft, C., Wall, K., Bizimana, J., Kilembe, W., Tichacek, A., Caliendo, A. M., Hunter, E., & Allen, S. (2011). Indeterminate and discrepant rapid HIV test results in couples' HIV testing and counselling centres in Africa. *J Int AIDS Soc*, *14*, 18. https://doi.org/10.1186/1758-2652-14-18

Boko, P. M., Ibikounle, M., Onzo-Aboki, A., Tougoue, J. J., Sissinto, Y., Batcho, W., Kinde-Gazard, D., & Kabore, A. (2016). Schistosomiasis and Soil Transmitted Helminths Distribution in Benin: A Baseline Prevalence Survey in 30 Districts. *PLoS One*, *11*(9), e0162798. https://doi.org/10.1371/journal.pone.0162798

Caffrey, C. R. (2015). Schistosomiasis and its treatment. *Future Medicinal Chemistry*, *7*(6), 675-676. https://doi.org/10.4155/fmc.15.27

Cameron, D. W., Simonsen, J. N., D'Costa, L. J., Ronald, A. R., Maitha, G. M., Gakinya, M. N., Cheang, M., Ndinya-Achola, J. O., Piot, P., Brunham, R. C., & et al. (1989). Female to male transmission of human immunodeficiency virus type 1: risk factors for seroconversion in men. *Lancet*, *2*(8660), 403-407. https://doi.org/10.1016/s0140-6736(89)90589-8

Cheever, A. W., Kamel, I. A., Elwi, A. M., Mosimann, J. E., & Danner, R. (1977). Schistosoma mansoni and S. haematobium infections in Egypt. II. Quantitative parasitological findings at necropsy. *The American journal of tropical medicine and hygiene*, *26*(4), 702-716. https://doi.org/10.4269/ajtmh.1977.26.702

Cioli, D., Pica-Mattoccia, L., Basso, A., & Guidi, A. (2014). Schistosomiasis control: praziquantel forever? *Molecular and Biochemical Parasitology*, *195*(1), 23-29. https://doi.org/https://doi.org/10.1016/j.molbiopara.2014.06.002

Downs, J. A., Dupnik, K. M., van Dam, G. J., Urassa, M., Lutonja, P., Kornelis, D., de Dood, C. J., Hoekstra, P., Kanjala, C., Isingo, R., Peck, R. N., Lee, M. H., Corstjens, P. L. A. M., Todd, J., Changalucha, J. M., Johnson, W. D., Jr., & Fitzgerald, D. W. (2017). Effects of schistosomiasis on susceptibility to HIV-1 infection and HIV-1 viral load at HIV-1 seroconversion: A nested case-control study. *PLoS Neglected Tropical Diseases*, *11*(9), e0005968. https://doi.org/10.1371/journal.pntd.0005968

Downs, J. A., Mguta, C., Kaatano, G. M., Mitchell, K. B., Bang, H., Simplice, H., Kalluvya, S. E., Changalucha, J. M., Johnson, W. D., Jr., & Fitzgerald, D. W. (2011). Urogenital schistosomiasis in women of reproductive age in Tanzania's Lake Victoria region. *The American journal of tropical medicine and hygiene*, *84*(3), 364-369. https://doi.org/10.4269/ajtmh.2011.10-0585

Edington, G. M., Nwabuebo, I., & Junaid, T. A. (1975). The pathology of schistosomiasis in Ibadan, Nigeria with special reference to the appendix, brain, pancreas and genital organs. *Trans R Soc Trop Med Hyg*, *69*(1), 153-156. https://doi.org/10.1016/0035-9203(75)90027-9

Gupta, S., Granich, R., & Williams, B. G. (2022). Update on treatment as prevention of HIV illness, death, and transmission: sub-Saharan Africa HIV financing and progress towards the 95–95–95 target. *Current Opinion in HIV and AIDS*, *17*(6). https://journals.lww.com/co-hivandaids/Fulltext/2022/11000/Update_on_treatment_as_prevention_of_HIV_illness,.7.aspx

Jourdan, P. M., Roald, B., Poggensee, G., Gundersen, S. G., & Kjetland, E. F. (2011). Increased Vascularity in Cervicovaginal Mucosa with Schistosoma haematobium Infection. *PLoS Neglected Tropical Diseases*, *5*(6), e1170. https://doi.org/10.1371/journal.pntd.0001170

Kayuni, S., Lampiao, F., Makaula, P., Juziwelo, L., Lacourse, E. J., Reinhard-Rupp, J., Leutscher, P. D. C., & Stothard, J. R. (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. https://doi.org/10.1016/j.parepi.2018.e00077

Kayuni, S. A., Alharbi, M. H., Makaula, P., Lampiao, F., Juziwelo, L., LaCourse, E. J., & Stothard, J. R. (2021). Male Genital Schistosomiasis Along the Shoreline of Lake Malawi: Baseline Prevalence and Associated Knowledge, Attitudes and Practices Among Local Fishermen in Mangochi District, Malawi. *Front Public Health*, *9*, 590695. https://doi.org/10.3389/fpubh.2021.590695

Kharsany, A. B. M., & Karim, Q. A. (2016). HIV Infection and AIDS in Sub-Saharan Africa: Current Status, Challenges and Opportunities. *The open AIDS journal*, *10*, 34-48. https://doi.org/10.2174/1874613601610010034

King, C. H., & Dangerfield-Cha, M. (2008). The unacknowledged impact of chronic schistosomiasis. *Chronic Illness*, *4*(1), 65-79. https://doi.org/10.1177/1742395307084407

Kjetland, E. F., Ndhlovu, P. D., Gomo, E., Mduluza, T., Midzi, N., Gwanzura, L., Mason, P. R., Sandvik, L., Friis, H., & Gundersen, S. G. (2006). Association between genital schistosomiasis and HIV in rural Zimbabwean women. *Aids*, *20*(4), 593-600. https://doi.org/10.1097/01.aids.0000210614.45212.0a

Kjetland, E. F., Norseth, H. M., Taylor, M., Lillebø, K., Kleppa, E., Holmen, S. D., Andebirhan, A., Yohannes, T. H., Gundersen, S. G., Vennervald, B. J., Bagratee, J., Onsrud, M., & Leutscher, P. D. (2014). Classification of the lesions observed in female genital schistosomiasis. *Int J Gynaecol Obstet*, *127*(3), 227-228. https://doi.org/10.1016/j.ijgo.2014.07.014

Kleppa, E., Ramsuran, V., Zulu, S., Karlsen, G. H., Bere, A., Passmore, J. A., Ndhlovu, P., Lillebø, K., Holmen, S. D., Onsrud, M., Gundersen, S. G., Taylor, M., Kjetland, E. F., & Ndung'u, T. (2014). Effect of female genital schistosomiasis and anti-schistosomal treatment on monocytes, CD4+ T-cells and CCR5 expression in the female genital tract. *PLoS One*, *9*(6), e98593. https://doi.org/10.1371/journal.pone.0098593

Laga, M., Manoka, A., Kivuvu, M., Malele, B., Tuliza, M., Nzila, N., Goeman, J., Behets, F., Batter, V., Alary, M., Heyward, W. L., Ryder, R. W., & Piot, P. (1993). Non-ulcerative sexually transmitted diseases as risk factors for HIV-1 transmission in women: results from a cohort study. *Aids*, *7*(1). https://journals.lww.com/aidsonline/Fulltext/1993/01000/Non_ulcerative_sexually_transmitted_diseases_as.15.aspx

Lang, R., Minion, J., & Wong, A. (2017). Hematospermia in a returned traveler. *Canadian Urological Association Journal*, *11*(1-2), E41-43. https://doi.org/10.5489/cuaj.3836

Leutscher, P., Ramarokoto, C.-E., Reimert, C., Feldmeier, H., Esterre, P., & Vennervald, B. J. (2000). Community-based study of genital schistosomiasis in men from Madagascar. *The Lancet*, *355*(9198), 117-118. https://doi.org/10.1016/S0140-6736(99)04856-4

Leutscher, P. D., Pedersen, M., Raharisolo, C., Jensen, J. S., Hoffmann, S., Lisse, I., Ostrowski, S. R., Reimert, C. M., Mauclere, P., & Ullum, H. (2005). Increased prevalence of leukocytes and elevated cytokine levels in semen from Schistosoma haematobium-infected individuals. *J Infect Dis*, *191*(10), 1639-1647. https://doi.org/10.1086/429334

Leutscher, P. D. C., van Dam, G. T. J., Reimert, C. M., Ramarakoto, C.-E., Deelder, A. M., & Ørnbjerg, N. (2008). Eosinophil Cationic Protein, Soluble Egg Antigen, Circulating Anodic Antigen, and Egg Excretion in Male Urogenital Schistosomiasis. *The American Journal of Tropical Medicine and Hygiene Am J Trop Med Hyg*, *79*(3), 422-426. https://doi.org/10.4269/ajtmh.2008.79.422

Lothe, A., Zulu, N., Øyhus, A. O., Kjetland, E. F., & Taylor, M. (2018). Treating schistosomiasis among South African high school pupils in an endemic area, a qualitative study. *BMC Infectious Diseases*, *18*(1), 239. https://doi.org/10.1186/s12879-018-3102-0

Mbabazi, P. S., Andan, O., Fitzgerald, D. W., Chitsulo, L., Engels, D., & Downs, J. A. (2011). Examining the Relationship between Urogenital Schistosomiasis and HIV Infection. *PLoS Neglected Tropical Diseases*, *5*(12), e1396. https://doi.org/10.1371/journal.pntd.0001396

Patel, P., Rose, C. E., Kjetland, E. F., Downs, J. A., Mbabazi, P. S., Sabin, K., Chege, W., Watts, D. H., & Secor, W. E. (2021). Association of schistosomiasis and HIV infections: A systematic review and meta-analysis. *Int J Infect Dis*, *102*, 544-553. https://doi.org/10.1016/j.ijid.2020.10.088

Plummer, F. A., Simonsen, J. N., Cameron, D. W., Ndinya-Achola, J. O., Kreiss, J. K., Gakinya, M. N., Waiyaki, P., Cheang, M., Piot, P., Ronald, A. R., & et al. (1991). Cofactors in male-female sexual transmission of human immunodeficiency virus type 1. *J Infect Dis*, *163*(2), 233-239. https://doi.org/10.1093/infdis/163.2.233

Ramarakoto, C. E., Leutscher, P. D. C., van Dam, G., & Christensen, N. O. (2008). Ultrasonographical findings in the urogenital organs in women and men infected with Schistosoma haematobium in northern Madagascar. *Transactions of The Royal Society*

of Tropical Medicine and Hygiene, *102*(8), 767-773. https://doi.org/10.1016/j.trstmh.2008.03.007

Rebbapragada, A., Wachihi, C., Pettengell, C., Sunderji, S., Huibner, S., Jaoko, W., Ball, B., Fowke, K., Mazzulli, T., Plummer, F. A., & Kaul, R. (2007). Negative mucosal synergy between Herpes simplex type 2 and HIV in the female genital tract. *Aids*, *21*(5), 589-598. https://doi.org/10.1097/QAD.0b013e328012b896

Roberts, L., Passmore, J.-A. S., Mlisana, K., Williamson, C., Little, F., Bebell, L. M., Walzl, G., Abrahams, M.-R., Woodman, Z., Abdool Karim, Q., & Abdool Karim, S. S. (2012). Genital Tract Inflammation During Early HIV-1 Infection Predicts Higher Plasma Viral Load Set Point in Women. *The Journal of Infectious Diseases*, *205*(2), 194-203. https://doi.org/10.1093/infdis/jir715

Shane, H. L., Verani, J. R., Abudho, B., Montgomery, S. P., Blackstock, A. J., Mwinzi, P. N. M., Butler, S. E., Karanja, D. M. S., & Secor, W. E. (2011). Evaluation of Urine CCA Assays for Detection of Schistosoma mansoni Infection in Western Kenya. *PLoS Neglected Tropical Diseases*, *5*(1), e951. https://doi.org/10.1371/journal.pntd.0000951

Stecher, C. W., Kallestrup, P., Kjetland, E. F., Vennervald, B., & Petersen, E. (2015). Considering treatment of male genital schistosomiasis as a tool for future HIV prevention: a systematic review. *International Journal of Public Health*, *60*(7), 839-848. https://doi.org/10.1007/s00038-015-0714-7

van der Werf, M. J., de Vlas, S. J., Brooker, S., Looman, C. W., Nagelkerke, N. J., Habbema, J. D., & Engels, D. (2003). Quantification of clinical morbidity associated with schistosome infection in sub-Saharan Africa. *Acta Trop*, *86*(2-3), 125-139. https://doi.org/10.1016/s0001-706x(03)00029-9

Wall, K. M., Kilembe, W., Nizam, A., Vwalika, C., Kautzman, M., Chomba, E., Tichacek, A., Sardar, G., Casanova, D., Henderson, F., Mulenga, J., Kleinbaum, D., & Allen, S. (2012). Promotion of couples' voluntary HIV counselling and testing in Lusaka, Zambia by influence network leaders and agents. *BMJ Open*, *2*(5), e001171. https://doi.org/10.1136/bmjopen-2012-001171

Wall, K. M., Kilembe, W., Vwalika, B., Dinh, C., Livingston, P., Lee, Y.-M., Lakhi, S., Boeras, D., Naw, H. K., Brill, I., Chomba, E., Sharkey, T., Parker, R., Shutes, E., Tichacek, A., Secor, W. E., & Allen, S. (2018). Schistosomiasis is associated with incident HIV transmission and death in Zambia. *PLoS Neglected Tropical Diseases*, *12*(12), e0006902. https://doi.org/10.1371/journal.pntd.0006902

Wall, K. M., Kilembe, W., Vwalika, B., Haddad, L. B., Hunter, E., Lakhi, S., Chavuma, R., Htee Khu, N., Brill, I., Vwalika, C., Mwananyanda, L., Chomba, E., Mulenga, J., Tichacek, A., & Allen, S. (2017). Risk of heterosexual HIV transmission attributable to sexually transmitted infections and non-specific genital inflammation in Zambian discordant couples, 1994-2012. *Int J Epidemiol*, *46*(5), 1593-1606. https://doi.org/10.1093/ije/dyx045

Wharton-Smith, A., Rassi, C., Batisso, E., Ortu, G., King, R., Endriyas, M., Counihan, H., Hamade, P., & Getachew, D. (2019). Gender-related factors affecting health seeking for neglected tropical diseases: findings from a qualitative study in Ethiopia. *PLoS Negl Trop Dis*, *13*(12), e0007840. https://doi.org/10.1371/journal.pntd.0007840