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Oumaima Kaabi Date

Associations between schistosome infection and incident HIV not mediated by STIs or vaginal dysbiosis in a longitudinal cohort of Zambian women

By

Oumaima Kaabi

Master of Public Health
Global Epidemiology

Kristin M Wall, MS, PhD
Committee Chair

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Oumaima Kaabi

Bachelor of Science
Virginia Commonwealth University
2016

Thesis Committee Chair: Kristin M Wall, MS, PhD

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Abstract

Associations between schistosome infection and incident HIV not mediated by STIs or vaginal dysbiosis in a longitudinal cohort of Zambian women

By Oumaima Kaabi

Introduction. Schistosomiasis affects over 240 million people worldwide. The species *S. haematobium* causes urogenital schistosomiasis and is most common in Zambia. Female genital schistosomiasis (FGS) is associated with increased risk of HIV, but the mechanism requires further elucidation. FGS is suspected to increase women's risk for sexually transmitted infections (STIs) and vaginal dysbiosis which are known HIV risk factors. In this analysis we test whether STIs, vaginal dysbiosis, and genital ulcers are mediators for the association between schistosomiasis and HIV risk in women who participated in a longitudinal cohort of heterosexual HIV serodiscordant couples in Zambia.

Methods. Data and samples were collected from a prospective cohort in Lusaka, Zambia from 1994-2009. Baseline demographic variables are presented using descriptive statistics. We use Cox survival models to evaluate whether genital inflammation, STIs (gonorrhea, chlamydia, trichomonas), bacterial vaginosis (BV), candida, and genital ulcers are mediators of the association between schistosomiasis and HIV by evaluating their association with schistosome-specific (SS) antibody status. Data are stratified by HIV status and SS antibody status.

Results. 50% of HIV+ women (n=596) had positive SWAP ELISA results at baseline compared with 55% of HIV- women (n=503). Schistosome infection was not significantly or meaningfully associated STIs, genital inflammation, bacterial vaginosis (BV), candida, or genital ulcers. SS+ women were less likely to be pregnant at baseline and more likely to have a partner who is also SS+. SS+/HIV+ women were more likely to be in HIV Stage III-IV than SS-/HIV+ women. In HIV+ women, SS+ status was associated with lower levels of unprotected sex and fewer incident pregnancies.

Discussion. The data from this cohort does not support the hypothesis that the association between FSG and HIV risk is mediated by STIs. This is one of the first studies to use data from a large, robust, observational cohort to evaluate this hypothesis. Future studies should examine other mechanisms that may explain the relationship between FGS and HIV including genital tract damage facilitating HIV entry or changes in immune markers leading to increased HIV risk. Understanding this pathway will inform more targeted interventions that can control the HIV epidemic in the region.

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Chapter I: Background and Literature Review

Schistosomiasis Epidemiology

Schistosomiasis is a disease caused by fresh water parasitic worms and can manifest in a variety of symptoms including anemia, stunned growth, and learning disabilities in children (Mbabazi et al., 2011). In countries where the parasite is endemic, infection typically occurs during childhood between ages 10-15, is more common in women than men, and is more common in rural setting compared to urban (Agnew-Blais et al., 2010). *S. haematobium* and *S. mansoni* are the two helminth species most common on the African continent, differentiated by presentation in the urogenital or intestinal tract, respectively (Kleppa et al., 2015). Urogenital schistosomiasis develops in 75% of people infected with the helminth species and is more likely to develop in women than in men (Mbabazi et al., 2011; Mutengo et al., 2009). 82 million women in Africa are estimated to be living with *Schistosoma* infections (Sturt et al., 2020). At the time of this study (1994-2009), disease burden among women in Lusaka was around 50-55% (Wall et al., 2018). Schistosomiasis can cause chronic disease and will last for decades if untreated (Mbabazi et al., 2011).

The presence of *S. haematobium* eggs in urogenital tracts causes a variety of symptoms and multiple a wide range of adverse health and social outcomes. Female genital schistosomiasis (FGS) is caused by *S. haematobium* infection occurring in the urogenital mucosa when eggs that are not excreted become lodged in the genital mucosa and form lesions (Engels et al., 2020). Schistosomiasis a whole is a neglected tropical disease and as a result, FGS has been understudied

even though it impacts 56 million women in sub-Saharan Africa alone (Engels et al., 2020; Sturt, Webb, Himschoot, et al., 2021).

FGS diagnosis typically results from the detection of *S. haematobium* eggs in the urogenital mucosa (Jourdan et al., 2011). Diagnosis is difficult due to challenges with the available diagnostic methods. Typically, an invasive colposcopy is done to confirm diagnose FGS (Mbabazi et al., 2011). Through a visual examination of the cervix, lesions, sandy patches, and abnormal blood vessels are typical presentation of FGS (Mbabazi et al., 2011). FGS diagnosis is associated with pelvic pain, altered cervical epithelium, genital inflammation, infertility, ectopic pregnancy, and stigma (Mbabazi et al., 2011; Sturt et al., 2020). There are limited studies focused on FGS, the mechanisms through which FGS causes the observed symptoms, or ways that it interacts with other co-occurring infections.

Schistosomiasis and HIV

Mapping of HIV endemic and schistosomiasis endemic countries produces extensive overlap (Mbabazi et al., 2011). Despite the geographical evidence suggesting that HIV/Schistosomiasis coinfection is common, there are a limited number of studies investigating the relationship between HIV and FGS and the implications of coinfection (Kleppa et al., 2015). Early research testing the hypothesis that these two infections were related was done in animal models and showed that acute schistosomiasis increases the risk of HIV acquisition (Chenine et al., 2008). Cross-sectional and cohort studies have since been used to show that there is three- to four-fold increase in HIV risk among women in various populations in sub-Saharan Africa (Mbabazi et al., 2011). These have shown that schistosomiasis infection is associated with an increased risk of HIV acquisition and transmission (Downs et al., 2017; Mbabazi et al., 2011; Sturt,

Webb, Phiri, et al., 2021; Wall et al., 2018). A longitudinal study in Zambia found that HIV positive women with detectable schistosome-specific antibodies (SS+) are more likely to transmit HIV to their HIV negative partner than schistosome antibody negative (SS-) women (Wall et al., 2018). In addition to transmission of HIV, women were more likely to acquire the virus from their partner in HIV discordant couples if they were SS+ than SS- and had increased progression from HIV detection to AIDS diagnosis and then death (Downs et al., 2017; Wall et al., 2018).

Mechanisms

There are a few working hypotheses as to how this relationship between HIV and schistosomiasis works, but it is thought to be causal as Schistosomiasis infection typically precedes HIV infection in the life course (Mbabazi et al., 2011; Sturt, Webb, Phiri, et al., 2021). Studies examining the mechanisms though which schistosomiasis infection results in higher rates of HIV transmission and acquisition suggest several different pathways. Genital inflammation leads to a measurable increase in the recruitment of CD4+ lymphocyte and macrophage, HIV target cells, to the genitals increases the risk of HIV acquisition (Jourdan et al., 2011; Kallestrup et al., 2005). Epithelial damage resulting from *S. haematobium* (SH+) infection also increases risk of HIV acquisition as the weakened structural integrity of the genital mucosa increases contact bleeding and facilitates virus entry (Mbabazi et al., 2011). Chronic schistosomiasis infection results in CD4+ T cells differentiating into Th2 T helper cells and the observed increase of viral replication within these cells when coupled with down-regulation of the Th-1 response typical to initial HIV infection may partially explain the increase in transmission and acquisition (Chenine et al., 2008; Maggi et al., 1994; Mbabazi et al., 2011). Another explanation is that higher leukocyte concentration in the genitals also increases risk of transmission of HIV from women to men

(Mbabazi et al., 2011). Increases in the pathogenesis of HIV in SH+ women may be attributed to the Th2-type immunologic response that is typical of extracellular parasitic infections (Paul & Zhu, 2010). Additionally, higher HIV viral loads have been observed in SH+ individuals, which is predictive of rapid disease progression (Mbabazi et al., 2011).

It has also been hypothesized that sexually transmitted infections (STIs) and other genital abnormalities in women may act as mediators in the pathway between schistosomiasis infection and HIV acquisition or transmission (Downs et al., 2017; Wall et al., 2018) (Fig 1). Previous data link various STIs and other causes of genital ulceration and inflammation (GUI) to HIV acquisition or transmission (Wall et al., 2017). The immune response to STIs like gonorrhea, chlamydia, trichomonas, syphilis, and herpes simplex virus type 2 (HSV-2) as well other causes of genital inflammation in women (like bacterial vaginosis (BV) & candida) recruits CD4 lymphocyte and macrophages that are HIV infection target cells to the genitalia, increasing HIV risk (Jourdan et al., 2011; Kallestrup et al., 2005; Mbabazi et al., 2011). The genital inflammation caused by these STIs has been shown to lead to higher HIV viral loads in women (Roberts et al., 2012). Inflammation coupled with disruptions in the cervical and vaginal epithelial barriers also caused by FGS also increase women's risk for HIV infection (Patel et al., 2021). However, to our knowledge, no study to date has evaluated the hypothesis that schistosomiasis infection increases HIV risk through a causal pathway mediated by STIs/GUIs (Fig 1).

Praziquantel Treatment

Praziquantel (PZQ) is an anti-parasitic chemotherapy that is currently the only drug used to control and treat both intestinal and urogenital (including FGS) schistosomiasis, as it has been since 1984 (Liu et al., 2011). As a very cost effective therapeutic, treating all those infected is

financially feasible given the at a cost of 40 cents per person (Hotez et al., 2009; Mbabazi et al., 2011). Most efficacy trials have been conducted on school-aged children as schistosomiasis is typically acquired during childhood but the drug has a 90% parasite egg reduction rate in all those infected, including adults (Zwang & Olliaro, 2014). While most people will experience at least one adverse side-effect like abdominal pain and nausea, the benefits of utilizing it as a treatment or a preventative prophylaxis dramatically outweigh the risk for harm and there is little evidence of widespread drug resistance (Doenhoff & Pica-Mattoccia, 2006; Liu et al., 2011; Zwang & Olliaro, 2014). The current World Health Organization (WHO) recommendation is to conduct mass drug administration of PZQ in endemic countries (Ndeffo Mbah et al., 2014). In addition to the benefits of reducing schistosomiasis morbidity and mortality, PZQ treatment has public health implications as a HIV control method (Ndeffo Mbah et al., 2014). Treatment of schistosomiasis results in reduced genital lesions and inflammation, which are through to increase HIV transmission and acquisition among women (Doenhoff & Pica-Mattoccia, 2006). The use of antiparasitic medication has also been shown to reduce the rate at which viral loads increase in SH/HIV coinfected individuals, and reduce viral load in semen, reducing HIV transmission from men to women (Mbabazi et al., 2011; Midzi et al., 2017). Praziquantel treatment before the age of 21 has also proven effective in reducing subfertility rates (Miller-Fellows et al., 2017). Despite the numerous benefits of PZQ treatment and evidence to support its use in decreasing the spread of HIV and the development of AIDS in sub-Saharan Africa, distribution and uptake have been low (Mbabazi et al., 2011).

Sociocultural Context and Ethical Considerations

In the context of gender identity and gender-specific societal roles, understanding the impact that schistosomiasis has on this population of women is critical for achieving health equity. FGS is a neglected tropical disease that disproportionally impacts women as their socially ascribed domestic duties (e.g., clothes washing, water fetching) leave them more likely to be exposed to *S. haematobium* contaminated water sources than men (Mubita-Ngoma, 2016; Sturt et al., 2020). This already vulnerable population therefore has a higher prevalence of schistosomiasis than men (Kjetland et al., 2010). In addition to the health impacts of infection (inflammation, infertility, pain, increase risk of HIV), the stigma associated with infection has wide reaching implications on women and girls' overall wellbeing (Sturt et al., 2020).

Schistosomiasis infection initially occurs during childhood in most cases (Ndeffo Mbah et al., 2014). For girls who are infected and develop FGS, the stigma can lead to significant effects on their health, social, and educational outcomes. FGS is not widely recognized, is difficult to diagnose, and some of its symptoms overlap with STI symptoms which often leads to its misdiagnosis as an STI (Mbabazi et al., 2011). For young girls in a conservative culture that views premarital sex negatively, this misdiagnoses of FGS as an STI leads to misplaced stigma that can lead to truancy and negative psychosocial effects (Bwalya, 2015). The association between FGS and stress incontinent can also lead girls to skip or stop attending school (Kjetland et al., 2008).

Lack of education and social support are predictors of poor health outcomes, especially when coupled with any potential mental health issues that may arise (Mubita-Ngoma, 2016). The stigmatization of infertility contributes to the social vulnerability of women (Inhorn & Patrizio, 2015). For women in low-resource settings, infertility effects physical health and psychosocial

wellbeing (Inhorn & Patrizio, 2015). Treating the underlying cause of the proportion of infertility cases caused by FGS can help women regain social roles that are important to them (Cui, 2010).

The perception of schistosomiasis as a predominantly rural disease causes disparities in research and resource allocation (Agnew-Blais et al., 2010). While the greater burden of disease is in hard-to-reach rural area, the lack of investment in diagnosing and investigating the impacts or differential risks of this disease among urban women only furthers the gender gap (Mbabazi et al., 2011). Innovation is required to minimize discomfort in the diagnosis of FGS and to conduct robust prospective epidemiologic studies that can establish causality while treating those with documented infection (Mbabazi et al., 2011). Understanding and controlling schistosomiasis becomes more important as global climate change increases environmental range for the helminth species (Kalinda et al., 2018; Mangal et al., 2008). The broad range of negative health effects of FGS demand an intersectional approach to research and interventions in order to holistically address the impacts of FGS.

Chapter II: Manuscript

Associations between schistosome infection and incident HIV not mediated by STIs or vaginal dysbiosis in a longitudinal cohort of Zambian women

By Oumaima Kaabi

Introduction. Schistosomiasis affects over 240 million people worldwide. The species *S. haematobium* causes urogenital schistosomiasis and is most common in Zambia. Female genital schistosomiasis (FGS) is associated with increased risk of HIV, but the mechanism requires further elucidation. FGS is suspected to increase women's risk for sexually transmitted infections (STIs) and vaginal dysbiosis which are known HIV risk factors. In this analysis we test whether STIs, vaginal dysbiosis, and genital ulcers are mediators for the association between schistosomiasis and HIV risk in women who participated in a longitudinal cohort of heterosexual HIV serodiscordant couples in Zambia.

Methods. Data and samples were collected from a prospective cohort in Lusaka, Zambia from 1994-2009. Baseline demographic variables are presented using descriptive statistics. We use Cox survival models to evaluate whether genital inflammation, STIs (gonorrhea, chlamydia, trichomonas), bacterial vaginosis (BV), candida, and genital ulcers are mediators of the association between schistosomiasis and HIV by evaluating their association with schistosome-specific (SS) antibody status. Data are stratified by HIV status and SS antibody status.

Results. 50% of HIV+ women (n=596) had positive SWAP ELISA results at baseline compared with 55% of HIV- women (n=503). Schistosome infection was not significantly or meaningfully associated STIs, genital inflammation, bacterial vaginosis (BV), candida, or genital ulcers. SS+ women were less likely to be pregnant at baseline and more likely to have a partner who is also SS+. SS+/HIV+ women were more likely to be in HIV Stage III-IV than SS-/HIV+ women. In HIV+ women, SS+ status was associated with lower levels of unprotected sex and fewer incident pregnancies.

Discussion. The data from this cohort does not support the hypothesis that the association between FSG and HIV risk is mediated by STIs. This is one of the first studies to use data from a large, robust, observational cohort to evaluate this hypothesis. Future studies should examine other mechanisms that may explain the relationship between FGS and HIV including genital tract damage facilitating HIV entry or changes in immune markers leading to increased HIV risk. Understanding this pathway will inform more targeted interventions that can control the HIV epidemic in the region.

Introduction

Schistosomiasis is one of the most neglected tropical diseases, affecting over 240 million people worldwide (Monde et al., 2016). *S. haematobium* and *S. mansoni* are the two most common schistosomiasis species endemic in Sub-Saharan Africa, causing urogenital and intestinal schistosomiasis, respectively. Schistosomiasis is waterborne and is transmitted via a parasitic worm that lives in freshwater. Cultural norms around who is responsible for fetching water leave women and children in the region especially vulnerable to infection as they are more often directly exposed to contaminated fresh water sources (Mubita-Ngoma, 2016; Sturt et al., 2020). Most Schistosomiasis infections occur during childhood and persist into adulthood (Ndeffo Mbah et al., 2014). There is growing concern over increasing transmission of Schistosomiasis due to the rise in global temperatures increasing the ecological range for the snail host (Kalinda et al., 2018; Mangal et al., 2008).

Female genital Schistosomiasis (FGS) is a specific form of urogenital schistosomiasis infection that has been inadequately addressed by both the medical field and policy makers (Engels et al., 2020). FGS can be difficult to diagnose, but typically results from the presence of S*. haematobium* eggs in the urogenital mucosa. FGS impacts 56 million women in Sub-Saharan Africa (Sturt, Webb, Phiri, et al., 2021). It is important to understand the wide range of health impacts schistosomiasis infection can have, especially in vulnerable populations. The long-term impacts of infection are varied and can include genital inflammation, ectopic pregnancy, infertility, and stigma (Sturt et al., 2020).

Notably, previously published data link FGS with increased risk for HIV acquisition (Patel et al., 2021; Wall et al., 2018). Additionally, a longitudinal cohort study in Zambia found that HIV

positive women with detectable schistosome antibodies are also more likely to transmit HIV to others and die earlier than schistosome antibody negative women (Wall et al., 2018). With the prevalence of HIV in Zambian women around 14.2% (UNAID, 2020), it is important to gain a better understanding of why FGS may increase HIV risk.

It has been hypothesized that sexually transmitted infections (STIs) and other genital abnormalities in women may act as mediators in the pathway between schistosomiasis infection and HIV acquisition or transmission (Downs et al., 2017; Wall et al., 2018) (Fig 1). Previous data link various STIs and other causes of genital ulceration and inflammation (GUI) to HIV acquisition or transmission (Wall et al., 2017). The immune response to STIs like gonorrhea, chlamydia, trichomonas, syphilis, and herpes simplex virus type 2 (HSV-2) as well other causes of genital inflammation in women (like bacterial vaginosis (BV) & candida) recruits CD4 lymphocyte and macrophages that are HIV infection target cells to the genitalia, increasing HIV risk (Jourdan et al., 2011; Kallestrup et al., 2005). The genital inflammation caused by these STIs has been shown to lead to higher HIV viral loads in women (Roberts et al., 2012). Inflammation coupled with disruptions in the cervical and vaginal epithelial barriers also caused by FGS also increase women's risk for HIV infection (Patel et al., 2021). However, to our knowledge, no study to date has evaluated the hypothesis that schistosomiasis infection increases HIV risk through a causal pathway mediated by STIs/GUIs (Fig 1).

Understanding the ways through which FGS infection increases HIV infections will allow for more effective treatment and prevention interventions. Currently the World Health Organization (WHO) recommends mass drug administration of praziquantel (PZQ), a relatively inexpensive anthelmintic, to control the Schistosomiasis in countries where it is endemic

(Mbabazi et al., 2011; Ndeffo Mbah et al., 2014). Previously published research has shown that treating Schistosomiasis could dramatically and cost-effectively decrease in HIV transmission (Ndeffo Mbah et al., 2014). If STIs and GUI mediate the relationship between Schistosomiasis and HIV risk, treatment, and prevention of STIs may also be a strategy to reduce HIV risk attributable to schistosomiasis.

In this retrospective analysis of a longitudinal cohort study of adult heterosexual couples discordant for HIV in Zambia, we test the hypothesis that the association between schistosomiasis and HIV risk is mediated by STI/GUIs. In Zambia, *S. haematobium* is endemic and causes the majority of schistosomiasis infections (Mutengo et al., 2009). Prevalence of *S. haematobium* infection is over 22% in Zambia, it is spread in both rural and urban areas, and many people remain vulnerable to infection (Sturt et al., 2020).

Methods

Study Participants

Informed consent was obtained for all enrolled participants following study approval by Emory University and University of Zambia's Institutional Review Boards. The study cohort consists of heterosexual couples with discordant HIV status enrolled in Lusaka, Zambia between 1994 and 2009. This analysis focuses on the female partner in the M+F- and M-F+ couples. Follow-up visits were conducted every 3 months. All couples enrolled participated in voluntary HIV risk reduction counseling and testing including group educational sessions, rapid HIV antibody testing, and joint post-test couples' counseling. Eligible couples who enrolled were provided with free care including family planning and regular STI testing and treatment. Couples were censored

from the study upon antiretroviral treatment initiation by the HIV+ partner, death of either partners, or dissolution of the partnership.

Demographic and Time-Varying Variables

Demographic data measured at baseline include age, years cohabitating, monthly income. Clinical characteristics measured at baseline include pregnancy status, fertility intentions, HIV stage, viral load of HIV positive partners, circumcision status of the male partner, and history of STIs. STI history refers to self-reported instances of gonorrhea, chlamydia, trichomonas, syphilis, and HSV-2 diagnosis within the past year. Serology testing via the highly sensitive test Focus Diagnostics HerpeSelect 2 ELISA IgG, was done at baseline to detect HSV-2 infection.

Time-varying covariates measured at each visit include family planning method used, condomless sex, and incident pregnancy. Microscopy and physical exams were also conducted longitudinally. Microscopy of vaginal wet mount swabs was used to detect trichomonas, bacterial vaginosis (BV), candida, and sperm. A rapid plasma regain (RPR) was used to test for syphilis with a Treponema pallidum hemagglutination assay to confirm the results if available. Genital ulceration including cervical/vaginal erosion or friability was determined by physical exam finding and/or participant self-report. A visual genital exam was conducted to detect abnormalities like endocervical discharge and inflammation.

Outcomes of interest: genital abnormalities

Our time-varying outcomes of interest included non-STI genital inflammation (defined as any genital inflammation noted on physical exam without a corresponding STI diagnosis), STI genital inflammation (defined as presence of endocervical discharge noted on physical exam

indicative of gonorrhea or chlamydia or trichomonas diagnosed by microscopy), BV, candida, bilateral inguinal adenopathy diagnoses on physical exam, or genital ulcer diagnosed on physical exam and/or participant self-report.

Exposure of interest: Schistosomiasis Antibodies

Serum and blood plasma samples were collected from all participants at the time of enrollment and stored in a repository at Emory University. For samples collected from 1994 to 2009, retrospective ELISA testing of plasma samples was done for antibodies to schistosome soluble worm antigen preparation (SWAP) using ELISA. This nested case-control design included all participants who seroconverted as cases and a random sample of those who did not seroconvert as controls. ELISA data was incomplete for some discordant couples. Assay control included a 1:3 serial dilution on each plate and a 4-parameter curve fitting model was used to assign values to each sample based on the standard curve. 25 units was chosen as the serum positive cutoff as that is three standard deviations above average anti-SWAP IgG in serum from egg negative controls in the US and Europe. A positive schistosomiasis result is defined as having a positive SWAP antibody response. To secure free PZQ treatment for those who retrospectively tested positive, a list of positive schistosome-specific antibody was sent to the Director of the Lusaka research site.

Statistical Data Analysis

Baseline demographic variables are presented using descriptive statistics stratified by HIV status and schistosomiasis ELISA antibody status. Counts and percentages are presented for categorical characteristics. Means, and standard deviations are presented for continuous variables, with differences evaluated using Chi-square or t-tests, respectively. Cox survival

models were used to calculate unadjusted associations between time varying outcomes of interest and SWAP ELISA results. Other time-varying factors were considered as potential confounders, including having unprotected sex with HIV discordant study partner within the last 3 months, presence of sperm on the vaginal wet mount swabs, and incident pregnancy. Report includes crude hazard ratios (cHRs), 95% confidence intervals (CIs), and two-tailed p-values are reported. All analyses were performed with SAS v9.4.

Results

We analyzed data from n=1105 women who had an average follow up time of two years and mean age of 28. 50% of HIV+ women (n=596) had positive SWAP ELISA results at baseline compared with 55% of HIV- women (n=503). There are no significant differences in mean age, years cohabiting with partner, and monthly household income between women who tested positive for schistosome-specific (SS) antibodies (SS+) and those who did not (SS-). Partners of SS+ women were also more like to be SS+ themselves.

Positive SWAP ELISA results are associated with lower instances of baseline pregnancy in both HIV+ women ($p<.0001$) and HIV- women ($p=0.033$) but were not associated with fertility intentions within the upcoming year. Among HIV+ women, presence of schistosomiasis antibodies at baseline is associated with HIV disease progression, with 43% of the women living with HIV and schistosomiasis antibody positive in stage III-IV compared with 29% of HIV+ women who were schistosomiasis antibody negative ($p=0.001$). Viral load, circumcision status of male partner, HSV-2 status, RPR status, and history of STI within the past year were not associated with schistosomiasis antibody status for HIV+ or HIV- women (Table 1).

There were no meaningful or statistically significant associations between baseline schistosome infection and time varying variables including genital inflammation (STI or non-STI), bacterial vaginosis (BV), candida, bilateral inguinal adenopathy, or genital ulcers. In HIV+ women, schistosomiasis antibody positive status was associated with lower levels of unprotected sex with study partner within the last 3 month and fewer incident pregnancies (Table 2).

Discussion

In this cohort of women residing in an urban environment, 52% of participants tested positive for schistosome-specific antibodies (SS+), which contradicts traditional thinking of schistosomiasis infection as a childhood condition most prominent in rural settings (Kalinda et al., 2018). HIV incidence in this cohort is also high at 7.5/100PY for women acquiring HIV from their partner and 5.3/100PY for women transmitting HIV to their partner (Wall et al., 2017). Previously published data showed that during follow-up, 92 of n=275 HIV- women acquired HIV from their HIV+ male partners and 70 of n=296 HIV+ women transmitted the virus to their HIV- partner (Wall et al., 2018). Given that this population has concurrently high SS+ prevalence and high HIV incidence, a better understanding of the association between schistosomiasis infection and HIV could lead to additional strategies to control the HIV epidemic in schistosomiasis endemic areas.

There are a few mechanisms through which schistosomiasis infection is thought to be associated with HIV acquisition and transmission. It has been shown in an animal model that schistosomiasis infection leaves the host more susceptible to HIV acquisition due to genital mucosal damage (Siddappa et al., 2011). In women specifically, the presence of *S. haematobium* eggs in genital tissues leads a fragile mucosa that is vulnerable to contact bleeding, increasing the

risk of HIV acquisition and transmission (Kleppa et al., 2015). The host's immune response to the schistosomiasis infection is also implicated due to the recruitment of CD4 T cells and other immune markers in response to the presence of the helminth (Chenine et al., 2008). Men who have been treated for schistosomiasis have been shown to have lower HIV viral loads in semen (Midzi et al., 2017). This supports the hypothesis that schistosomiasis infection may be increasing the transmission of HIV through higher viral loads in semen.

While it was hypothesized that time varying STIs/GUIs can be mediators for the relationship between schistosomiasis and HIV (Downs et al., 2017; Wall et al., 2018), we did not observe that STIs/GUIs were important mediators. In this cohort of 1105 women with an average follow-up time of 2 years, data indicate that the observed association between schistosomiasis antibody status and incident HIV infection are not mediated by genital abnormalities including non-STI and STI inflammation, BV, candida, bilateral inguinal adenopathy, and genital ulcers ($p>0.05$). The presence or absence of one of the listed time-varying STIs did not impact these women's risk of acquiring or transmitting HIV based on their baseline SS antibody status at baseline.

Among HIV+ women, SS antibody positive women were slightly less likely than SS antibody negative women to have had unprotected sex with their study partner in the last 3 months ($p=0.003$), and incident pregnancy was lower for SS antibody positive women than SS antibody negative women ($p=0.005$) despite there being no difference in fertility intentions between the two groups. This finding supports the link between schistosomiasis infection and infertility. FGS infection is marked by the presence of Schistosoma ova in the genital tract leading to inflammation and development of scar tissue, symptoms that are associated with infertility

and complicated pregnancies (Kjetland et al., 2010). This finding of lower incident pregnancy in HIV+/SS+ women is consistent with existing literature given that HIV+ women also have higher rates of infertility than their HIV- counterparts (Santulli et al., 2011). In general, infertility in sub-Saharan Africa impacts around 30% of the population and a significant proportion may be due to the presence of *S. haematobium* (Miller-Fellows et al., 2017). For women in low-resource settings, infertility not only effects their physical health, but can also has negative psychosocial implications (Inhorn & Patrizio, 2015).

In the context of gender identity and gender specific societal roles, understanding the impact that schistosomiasis has on this population of women is critical for achieving health equity. This is especially important as women's socially ascribed domestic duties (e.g., clothes washing, water fetching) put them at higher risks of exposure to *S. haematobium* contaminated water (Kjetland et al., 2010). Due to the stigmatization of infertility, treating the underlying cause of the proportion of infertility cases caused by FGS can help women regain social roles that are important to them (Cui, 2010). The argument for increasing mass drug administration efforts to treat schistosomiasis are therefore multifold. PZQ treatment before the age of 21 has proven effective in reducing subfertility rates (Miller-Fellows et al., 2017). When coupled with a model predicting that HIV transmission can be reduced by 20% over 20 years following mass drug administration to treat schistosomiasis, the argument for continuing research on the association between schistosomiasis and HIV is compelling (Ndeffo Mbah et al., 2014).

There are limitations to this study. ELISA testing of participant samples was done retrospectively for a subset of the total cohort. We did not use gold standard tests for detecting active infection (Shane et al., 2011) but rather antibody testing identified individuals with both

current and past infection. For future studies, confirmation of FGS via colposcopy or cervical biopsy to detect schistosome eggs is recommended (Wall et al., 2018). Time-varying measures of schistosome antibody levels rather than only baseline measures could also help inform how the level of infection changes over time. Additional information on proximity to infected water source and prior treatment (PZQ and assisted reproductive treatments) can help us further understand the nuances of schistosomiasis infection on HIV acquisition and impacts on infertility. Not all RPR results were confirmed via Treponema pallidum hemagglutination assay and may be a potential source of bias.

Chapter III: Public Health Implications and Future Directions

Schistosomiasis as a neglected tropical disease has not been adequately addressed through research or policy decisions (Patel et al., 2021). The implementation of the World Health Organization's (WHO) current recommendation of mass drug administration of praziquantel (PZQ) has broad public health implications that can impact over 240 million people worldwide (Monde et al., 2016; Ndeffo Mbah et al., 2014). Female genital Schistosomiasis (FGS) is a specific form of urogenital schistosomiasis resulting from the presence of *S. haematobium* eggs in the urogenital mucosa and impacts 56 million women in Sub-Saharan Africa (Sturt, Webb, Phiri, et al., 2021). Given the established association between FGS and increased risk of HIV acquisition and transmission, mass drug administration of PZQ can be a significant tool in the effort to control the HIV epidemic in Sub-Saharan Africa (Liu et al., 2011). An effective public health campaign can go a long way in educating the lay public about schistosomiasis, its symptoms, and encouraging seeking medical attention when infection is suspected. Programs for testing local fresh water sources may be an effective method for establishing zones where regular testing is recommended and determining effective locations for centers for PZQ distribution. As climate change causes higher temperature and changes rain/drought patterns and as development leads to more damns and irrigation projects, the geographical distribution of the helminth may shift and regular testing can help track this (De Leo et al., 2020). This would also allow future studies to have information regarding proximity to infected water source. The difficulty in diagnosing FGS and distinguishing the way it presents in the urogenital tracts from STIs denotes the importance of specific training for health care professionals on recognizing schistosomiasis and FGS.

Investment in addressing the impacts of schistosomiasis infection has widespread impacts on women as they are the most impacted population. Disparities in infection and health outcomes between men and women arise from differential exposure to contaminated water sources due to socially ascribed domestic duties (e.g., clothes washing, water fetching) (Kjetland et al., 2010). While the cultural values themselves should not be a target for epidemiologic public health intervention, understanding the socio-cultural influence on exposure risks and health outcomes will result in recommendations that are more likely to succeed. Addressing the multitude of health effects that schistosomiasis infection has can help women lead healthy lives and regain social roles that are important to them (Cui, 2010). Infertility in sub-Saharan Africa impacts around 30% of the population and a significant proportion may be due to the presence of *S. haematobium* (Miller-Fellows et al., 2017). Treating this infection will begin the process of addressing the negative psychosocial implications that arise from infertility resulting from FGS (Inhorn & Patrizio, 2015). More girls who may have experienced stigma from an STI misdiagnosis or stress incontinence will be able to continue their education and preventative treatment would prevent others from facing those challenges. Increase levels of education would have positive downstream health effects (Mubita-Ngoma, 2016).

Future studies should include be informed with the sociocultural context of the specified population. Further research should be done to investigate the pathways through which schistosomiasis infection increases risk of HIV infection to and transmission to elucidate the underlying mechanisms. This includes studies of cytokine and chemokine recruitment and activation through different phases of infection. Confirmation of FGS via colposcopy or cervical biopsy to detect lesions caused by schistosome eggs is recommended (WHO, 2020). Time-varying

measures of schistosome antibody levels rather than only baseline measures could also help inform how the level of infection changes over time. Future studies should also consider the impact of climate change on the range of infections (Kalinda et al., 2018).

Tables and Figures

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

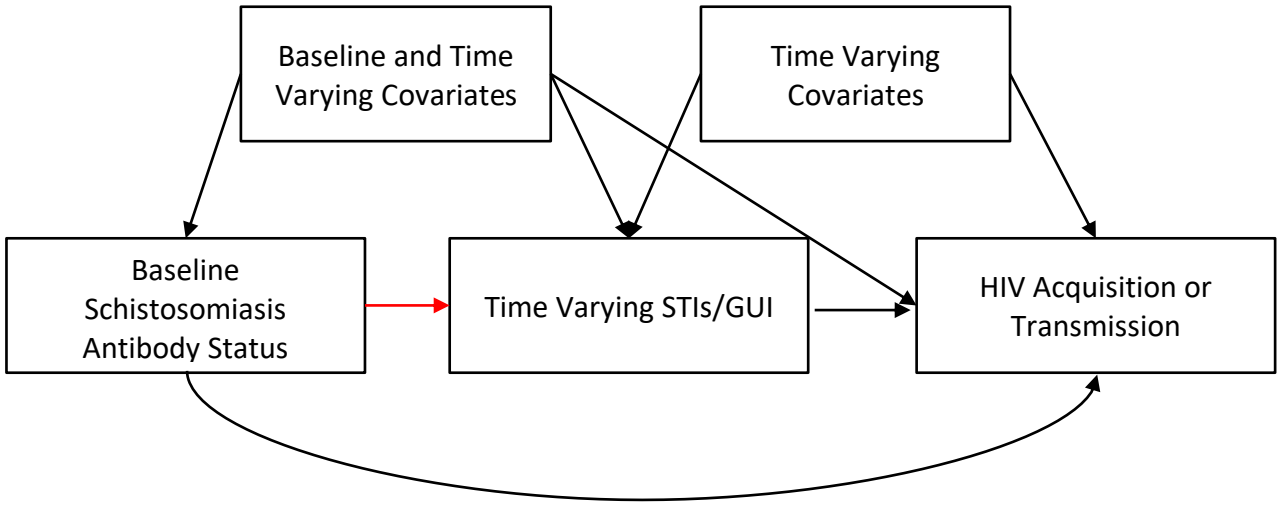

Table 1. Descriptive analyses and associations between baseline covariates by schistosome-specific (SS) antibody status among Zambian HIV+ and HIV- women

	HIV+ Women (N=596)					HIV- Women (N=503)				
	SS antibody positive (N=296)		SS antibody negative (N=300)		p-value	SS antibody positive (N = 275)		SS antibody negative (N=228)		p-value
	N	%	N	%		N	%	N	%	
Demographics										
Age (mean, SD)*	28.2	7.3	28.3	6.9	0.788	27.6	7.0	26.6	6.9	0.097
Years cohabiting (mean, SD)	6.2	7.2	5.6	5.8	0.222	7.6	6.5	7.4	6.7	0.740
Monthly household income (USD, mean, SD)***	60.2	76.5	71.3	107.1	0.147	59.8	78.6	67.1	70.3	0.282
Family planning characteristics										
Baseline pregnancy					<0.001					0.033
Yes	28	9%	64	21%		33	12%	43	19%	
No	268	91%	236	79%		242	88%	185	81%	
Fertility intentions					0.131					0.750
Yes, next year	11	17%	35	24%		13	33%	15	26%	
Yes, but not next year	12	19%	39	27%		8	20%	14	24%	
Don't know/No	41	64%	71	49%		19	48%	29	50%	
Clinical characteristics										
HIV stage, if positive					<0.001					
Stage I	84	28%	120	40%						
Stage II	84	28%	93	31%						
Stage III-IV	128	43%	87	29%						
VL, if positive (log10 copies/ml)*	4.6	0.9	4.4	0.9	0.094					
Circumcised male partner**					0.317					0.515
Yes	47	16%	39	13%		22	8%	22	10%	
No	249	84%	261	87%		253	92%	206	90%	
HSV-2 status					0.912					0.601
Positive	200	88%	167	87%		160	78%	101	74%	
Negative	17	7%	15	8%		29	14%	24	18%	
Indeterminate	11	5%	11	6%		15	7%	12	9%	
Past year history of any STI					0.559					0.844
Yes	136	46%	145	48%		96	35%	77	34%	
No	160	54%	155	52%		179	65%	149	66%	
RPR status of woman					0.975					0.155
Positive	2	1%	2	1%		5	2%	1	0%	
Negative	286	99%	295	99%		269	98%	227	100%	
SS antibody status of partner					0.002					0.004
Positive	188	76%	97	61%		197	76%	132	63%	
Negative	60	24%	62	39%		63	24%	76	37%	

Table 1 index

USD: United States Dollar; STI: sexually transmitted infection; OR: odds ratio; CI: confidence interval; HSV-2: herpes simplex virus 2; VL: viral load; SS: schistosome-specific
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
Variables not associated with the outcome (not tabled): contraception at baseline, male literacy, past year history of STI,

Not associated and not shown: Literacy in Nyanja

Table 2. Association between schistosome-specific (SS) antibody status and time-varying outcomes among HIV+ Zambian women

	HIV+ Women							
	SS antibody positive		SS antibody negative		cHR	95%CI		p-value
	N intervals	%	N intervals	%				
Time-varying outcomes of interest								
Non-STI genital inflammation					1.05	0.89	1.23	0.572
Yes	560	24%	510	13%				
No	1756	76%	3501	87%				
STI genital inflammation					1.08	0.82	1.43	0.586
Yes	234	10%	250	6%				
No	2082	90%	3761	94%				
BV					1.17	0.93	1.48	0.192
Yes	393	23%	312	14%				
No	1338	77%	1857	86%				
Candida					0.96	0.74	1.25	0.773
Yes	278	19%	257	20%				
No	1210	81%	1008	80%				
Bilateral inguinal adenopathy					1.07	0.83	1.37	0.599
Yes	228	14%	205	13%				
No	1377	86%	1397	87%				
Genital ulcer					1.10	0.86	1.41	0.446
Yes	376	16%	349	9%				
No	1913	84%	3551	91%				
Other time-varying (possible confounders)								
Any unprotected sex with study partner in last 3 months					0.82	0.72	0.94	0.003
Yes	860	37%	1565	40%				
No	1440	63%	2335	60%				
Sperm present on vaginal swab wet mount					0.92	0.70	1.20	0.534
Yes	249	13%	310	8%				
No	1669	87%	3349	92%				
Incident pregnancy					0.63	0.46	0.87	0.005
Yes	147	7%	325	9%				
No	2006	93%	3122	91%				

SS: schistosome-specific ; STI: sexually transmitted infection; cHR: crude hazard ratio; CI: confidence interval

Table 3. Association between schistosome-specific (SS) antibody status and time-varying outcomes among HIV- Zambian women

	HIV- Women							
	SS antibody positive		SS antibody negative		cHR	95%CI		p-value
	N intervals	%	N intervals	%				
Time-varying outcomes of interest								
Non-STI genital inflammation					0.96	0.82	1.13	0.625
Yes	462	25%	414	23%				
No	1352	75%	1403	77%				
STI genital inflammation					1.01	0.71	1.43	0.958
Yes	141	8%	128	7%				
No	1673	92%	1689	93%				
BV					1.04	0.78	1.37	0.808
Yes	252	16%	200	15%				
No	1278	84%	1151	85%				
Candida					0.87	0.69	1.11	0.268
Yes	267	19%	260	22%				
No	1153	81%	931	78%				
Bilateral inguinal adenopathy					0.73	0.48	1.09	0.123
Yes	78	5%	88	7%				
No	1397	95%	1243	93%				
Genital ulcer					1.08	0.75	1.55	0.687
Yes	173	10%	135	7%				
No	1641	90%	1678	93%				
Other time-varying (possible confounders)								
Any unprotected sex with study partner in last 3 months					0.89	0.77	1.03	0.103
Yes	669	37%	744	41%				
No	1143	63%	1066	59%				
Sperm present on vaginal swab wet mount					0.76	0.56	1.03	0.076
Yes	186	13%	237	15%				
No	1225	87%	1339	85%				
Incident pregnancy					0.90	0.69	1.17	0.427
Yes	180	10%	193	11%				
No	1576	90%	1550	89%				

SS: schistosome-specific ; STI: sexually transmitted infection; cHR: crude hazard ratio; CI: confidence interval

References

Agnew-Blais, J., Carnevale, J., Gropper, A., Shilika, E., Bail, R., & Ngoma, M. (2010, Aug). Schistosomiasis haematobium prevalence and risk factors in a school-age population of peri-urban Lusaka, Zambia. *J Trop Pediatr, 56*(4), 247-253. https://doi.org/10.1093/tropej/fmp106

Bwalya, L. (2015). *Assessing the Impact of re-entry policy in Zambian education system: A Case of Chongwe District* Mulungushi University]. SSRN.

Chenine, A. L., Shai-Kobiler, E., Steele, L. N., Ong, H., Augostini, P., Song, R., Lee, S. J., Autissier, P., Ruprecht, R. M., & Secor, W. E. (2008, Jul 23). Acute Schistosoma mansoni infection increases susceptibility to systemic SHIV clade C infection in rhesus macaques after mucosal virus exposure. *PLoS Negl Trop Dis, 2*(7), e265. https://doi.org/10.1371/journal.pntd.0000265

Cui, W. (2010, Dec 1). Mother or nothing: the agony of infertility. *Bull World Health Organ, 88*(12), 881-882. https://doi.org/10.2471/BLT.10.011210

De Leo, G. A., Stensgaard, A.-S., Sokolow, S. H., N'Goran, E. K., Chamberlin, A. J., Yang, G.-J., & Utzinger, J. (2020). Schistosomiasis and climate change. *Bmj*. https://doi.org/10.1136/bmj.m4324

Doenhoff, M. J., & Pica-Mattoccia, L. (2006, 2006/04/01). Praziquantel for the treatment of schistosomiasis: its use for control in areas with endemic disease and prospects for drug resistance. *Expert Review of Anti-infective Therapy, 4*(2), 199-210. https://doi.org/10.1586/14787210.4.2.199

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., Todd, J., Changalucha, J. M., Johnson, W. D., Jr., & Fitzgerald, D. W. (2017, Sep). Effects of schistosomiasis on susceptibility to HIV-1 infection and HIV-1 viral load at HIV-1 seroconversion: A nested case-control study. *PLoS Negl Trop Dis, 11*(9), e0005968. https://doi.org/10.1371/journal.pntd.0005968

Engels, D., Hotez, P. J., Ducker, C., Gyapong, M., Bustinduy, A. L., Secor, W. E., Harrison, W., Theobald, S., Thomson, R., Gamba, V., Masong, M. C., Lammie, P., Govender, K., Mbabazi, P. S., & Malecela, M. N. (2020, Sep 1). Integration of prevention and control measures for female genital schistosomiasis, HIV and cervical cancer. *Bull World Health Organ, 98*(9), 615-624. https://doi.org/10.2471/BLT.20.252270

Hotez, P. J., Fenwick, A., & Kjetland, E. F. (2009, May 26). Africa's 32 cents solution for HIV/AIDS. *PLoS Negl Trop Dis, 3*(5), e430. https://doi.org/10.1371/journal.pntd.0000430

Inhorn, M. C., & Patrizio, P. (2015, Jul-Aug). Infertility around the globe: new thinking on gender, reproductive technologies and global movements in the 21st century. *Hum Reprod Update, 21*(4), 411-426. https://doi.org/10.1093/humupd/dmv016

Jourdan, P. M., Holmen, S. D., Gundersen, S. G., Roald, B., & Kjetland, E. F. (2011, Dec). HIV target cells in Schistosoma haematobium-infected female genital mucosa. *Am J Trop Med Hyg, 85*(6), 1060-1064. https://doi.org/10.4269/ajtmh.2011.11-0135

Kalinda, C., Chimbari, M. J., & Mukaratirwa, S. (2018, Apr 30). Schistosomiasis in Zambia: a systematic review of past and present experiences. *Infect Dis Poverty, 7*(1), 41. https://doi.org/10.1186/s40249-018-0424-5

Kallestrup, P., Zinyama, R., Gomo, E., Butterworth, A. E., Mudenge, B., van Dam, G. J., Gerstoft, J., Erikstrup, C., & Ullum, H. (2005, 12/2005). Schistosomiasis and HIV-1 Infection in Rural Zimbabwe: Effect of Treatment of Schistosomiasis on CD4 Cell Count and Plasma HIV-1 RNA Load. *The Journal of Infectious Diseases*(11), 6. https://doi.org/10.1086/497696

Kjetland, E. F., Kurewa, E. N., Mduluza, T., Midzi, N., Gomo, E., Friis, H., Gundersen, S. G., & Ndhlovu, P. D. (2010, Sep). The first community-based report on the effect of genital Schistosoma haematobium infection on female fertility. *Fertil Steril, 94*(4), 1551-1553. https://doi.org/10.1016/j.fertnstert.2009.12.050

Kjetland, E. F., Kurewa, E. N., Ndhlovu, P. D., Midzi, N., Gwanzura, L., Mason, P. R., Gomo, E., Sandvik, L., Mduluza, T., Friis, H., & Gundersen, S. G. (2008, Dec). Female genital schistosomiasis--a differential diagnosis to sexually transmitted disease: genital itch and vaginal discharge as indicators of genital Schistosoma haematobium morbidity in a cross-sectional study in endemic rural Zimbabwe. *Trop Med Int Health, 13*(12), 1509-1517. https://doi.org/10.1111/j.1365-3156.2008.02161.x

Kleppa, E., Klinge, K. F., Galaphaththi-Arachchige, H. N., Holmen, S. D., Lillebo, K., Onsrud, M., Gundersen, S. G., Taylor, M., Ndhlovu, P., & Kjetland, E. F. (2015). Schistosoma haematobium infection and CD4+ T-cell levels: a cross-sectional study of young South African women. *PLoS One, 10*(3), e0119326. https://doi.org/10.1371/journal.pone.0119326

Liu, R., Dong, H.-F., Guo, Y., Zhao, Q.-P., & Jiang, M.-S. (2011). Efficacy of praziquantel and artemisinin derivatives for the treatment and prevention of human schistosomiasis: a systematic review and meta analysis. *Parasites & Vectors, 4*, 17.

Maggi, E., Mazzetti, M., Ravina, A., Annunziato, F., De Carli, M., Piccinni, M. P., Manetti, R., Carbonari, M., Pesce, A. M., Del Prete, G., & Romagnani, S. (1994). Ability of HIV to Promote a TH1 to TH0 Shift and to Replicate Preferentially in TH2 and TH0 Cells. *Science, 265*, 6.

Mangal, T. D., Paterson, S., & Fenton, A. (2008, Jan 16). Predicting the impact of long-term temperature changes on the epidemiology and control of schistosomiasis: a mechanistic model. *PLoS One, 3*(1), e1438. https://doi.org/10.1371/journal.pone.0001438

Mbabazi, P. S., Andan, O., Fitzgerald, D. W., Chitsulo, L., Engels, D., & Downs, J. A. (2011, Dec). Examining the relationship between urogenital schistosomiasis and HIV infection. *PLoS Negl Trop Dis, 5*(12), e1396. https://doi.org/10.1371/journal.pntd.0001396

Midzi, N., Mduluza, T., Mudenge, B., Foldager, L., & Leutscher, P. D. C. (2017, Fall). Decrease in Seminal HIV-1 RNA Load After Praziquantel Treatment of Urogenital Schistosomiasis Coinfection in HIV-Positive Men-An Observational Study. *Open Forum Infect Dis, 4*(4), ofx199. https://doi.org/10.1093/ofid/ofx199

Miller-Fellows, S. C., Howard, L., Kramer, R., Hildebrand, V., Furin, J., Mutuku, F. M., Mukoko, D., Ivy, J. A., & King, C. H. (2017, Nov). Cross-sectional interview study of fertility, pregnancy, and urogenital schistosomiasis in coastal Kenya: Documented treatment in childhood is associated with reduced odds of subfertility among adult women. *PLoS Negl Trop Dis, 11*(11), e0006101. https://doi.org/10.1371/journal.pntd.0006101

Monde, C., Syampungani, S., & van den Brink, P. J. (2016, Jun). Natural and human induced factors influencing the abundance of Schistosoma host snails in Zambia. *Environ Monit Assess, 188*(6), 370. https://doi.org/10.1007/s10661-016-5351-y

Mubita-Ngoma, C. A. (2016). Factors Influencing Women's Optimum Health in Zambia. *Journal of Healthcare Communications, 01*(04). https://doi.org/10.4172/2472-1654.100030

Mutengo, M. M., Mudenda, V., Mwansa, J. C., Kaonga, K., Sianongo, S., Wamulume, H. I., & Shinondo, C. J. (2009, 2009). Presence of Schistosomiasis in Genital Biopsies from patients at the University Teaching Hospital in Lusaka, Zambia. *Medical Journal of Zambia, 36*(3), 5.

Ndeffo Mbah, M. L., Gilbert, J. A., & Galvani, A. P. (2014, Jun). Evaluating the potential impact of mass praziquantel administration for HIV prevention in Schistosoma haematobium high-risk communities. *Epidemics, 7*, 22-27. https://doi.org/10.1016/j.epidem.2014.04.002

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, Jan). 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

Paul, W. E., & Zhu, J. (2010, Apr). How are T(H)2-type immune responses initiated and amplified? *Nat Rev Immunol, 10*(4), 225-235. https://doi.org/10.1038/nri2735

Roberts, L., Passmore, J. A., Mlisana, K., Williamson, C., Little, F., Bebell, L. M., Walzl, G., Abrahams, M. R., Woodman, Z., Abdool Karim, Q., & Abdool Karim, S. S. (2012, Jan 15).

Genital tract inflammation during early HIV-1 infection predicts higher plasma viral load set point in women. *J Infect Dis, 205*(2), 194-203. https://doi.org/10.1093/infdis/jir715

Santulli, P., Gayet, V., Fauque, P., Chopin, N., Dulioust, E., Wolf, J. P., Chapron, C., & de Ziegler, D. (2011, Feb). HIV-positive patients undertaking ART have longer infertility histories than age-matched control subjects. *Fertil Steril, 95*(2), 507-512. https://doi.org/10.1016/j.fertnstert.2010.09.018

Shane, H. L., Verani, J. R., Abudho, B., Montgomery, S. P., Blackstock, A. J., Mwinzi, P. N., Butler, S. E., Karanja, D. M., & Secor, W. E. (2011, Jan 25). Evaluation of urine CCA assays for detection of Schistosoma mansoni infection in Western Kenya. *PLoS Negl Trop Dis, 5*(1), e951. https://doi.org/10.1371/journal.pntd.0000951

Siddappa, N. B., Hemashettar, G., Shanmuganathan, V., Semenya, A. A., Sweeney, E. D., Paul, K. S., Lee, S. J., Secor, W. E., & Ruprecht, R. M. (2011, Aug). Schistosoma mansoni enhances host susceptibility to mucosal but not intravenous challenge by R5 Clade C SHIV. *PLoS Negl Trop Dis, 5*(8), e1270. https://doi.org/10.1371/journal.pntd.0001270

Sturt, A. S., Webb, E. L., Himschoot, L., Phiri, C. R., Mapani, J., Mudenda, M., Kjetland, E. F., Mweene, T., Levecke, B., van Dam, G. J., Corstjens, P., Ayles, H., Hayes, R. J., van Lieshout, L., Hansingo, I., Francis, S. C., Cools, P., & Bustinduy, A. L. (2021, Sep). Association of Female Genital Schistosomiasis With the Cervicovaginal Microbiota and Sexually Transmitted Infections in Zambian Women. *Open Forum Infect Dis, 8*(9), ofab438. https://doi.org/10.1093/ofid/ofab438

Sturt, A. S., Webb, E. L., Phiri, C. R., Mudenda, M., Mapani, J., Kosloff, B., Cheeba, M., Shanaube, K., Bwalya, J., Kjetland, E. F., Francis, S. C., Corstjens, P., van Dam, G. J., van Lieshout, L., Hansingo, I., Ayles, H., Hayes, R. J., & Bustinduy, A. L. (2021, Jul). Female Genital Schistosomiasis and HIV-1 Incidence in Zambian Women: A Retrospective Cohort Study. *Open Forum Infect Dis, 8*(7), ofab349. https://doi.org/10.1093/ofid/ofab349

Sturt, A. S., Webb, E. L., Phiri, C. R., Mweene, T., Chola, N., van Dam, G. J., Corstjens, P., Wessels, E., Stothard, J. R., Hayes, R., Ayles, H., Hansingo, I., van Lieshout, L., & Bustinduy, A. L. (2020, Jul). Genital self-sampling compared with cervicovaginal lavage for the diagnosis of female genital schistosomiasis in Zambian women: The BILHIV study. *PLoS Negl Trop Dis, 14*(7), e0008337. https://doi.org/10.1371/journal.pntd.0008337

UNAID. (2020). Country Progress Report - Zambia: Global AIDS Monitoring.

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, Dec). Schistosomiasis is associated with incident HIV transmission and death in Zambia. *PLoS Negl Trop Dis, 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, Oct 1). 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

WHO. (2020). Female Genital Schistosomiasis: A pocket atlas for clinical health-care professionals.

Zwang, J., & Olliaro, P. L. (2014). Clinical efficacy and tolerability of praziquantel for intestinal and urinary schistosomiasis-a meta-analysis of comparative and non-comparative clinical trials. *PLoS Negl Trop Dis, 8*(11), e3286. https://doi.org/10.1371/journal.pntd.0003286