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Prevalence and Factors Associated With Hepatitis B and C Co-Infection Among HIV-1-Infected Patients in Kenya

Duncan Ndegwa Maina,^a Andrew Kimanga Nyerere,^a Ruth Wambui Gicho,^a Joseph Maina Mwangi,^b Raphael Wekesa Lihana^b

^aCollege of Health Sciences, Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya; ^bKenya Medical Research Institute, Nairobi, Kenya
Correspondence to Raphael Lihana (lihanaraphael@gmail.com).

ABSTRACT

Background: Hepatitis B virus (HBV) and hepatitis C virus (HCV) are among the most chronic viral infections worldwide. Co-infections with HBV and HCV have become increasingly common among people living with HIV, resulting in a growing public health concern. The primary aim of our study was to determine the prevalence of HBV and HCV and their associated factors among HIV-1-infected patients attending the Ngong Sub-County Hospital comprehensive care clinic.

Methods: After providing consent, a 5 mL blood sample was collected from each study participant visiting the comprehensive care clinic. The blood was screened for hepatitis B surface antigen and HCV antibodies using chemiluminescence immunoassay test according to the manufacturer's instructions. The CD4 T-cell counts were determined using FACSCalibre machine, while HIV-1 viral load was determined using the Abbott m2000rt System according to the manufacturer's instructions. A questionnaire was used to collect sociodemographic information and data on factors associated with HBV and HCV co-infections.

Results: One hundred and ninety HIV-1-infected patients participated in this study: 150 (78.9%) women and 40 (21.1%) men. In the overall study population, the prevalence of HBV co-infection was 5.8% (95% CI, 2.6%–8.9%) and of HCV co-infection was 4.2% (95% CI, 1.6%–7.4%). However, no individual was co-infected with all 3 viruses. HCV was associated with antiretroviral treatment (OR 0.2; 95% CI, 0.0–0.8; $P=0.036$), while HBV showed a significant association with condom usage (OR 0.3; 95% CI, 0.1–0.9; $P=0.039$) and median viral load.

Conclusions: A high prevalence of HIV/HBV and HIV/HCV co-infection was reported in this study, suggesting that HIV-infected patients should be routinely screened for HBV and HCV infections, and preventive and control measures should be put in place that include public education on HBV and HCV infections.

BACKGROUND

Human immunodeficiency virus (HIV), hepatitis B virus (HBV) and hepatitis C virus (HCV) are among the most chronic viral pathogens of major public health concern worldwide.¹ More than 250 million people infected with HBV have developed chronic HBV infection, which has resulted in 800,000 HBV-related deaths annually.^{2–5} More than 70 million people have developed chronic HCV infection globally.^{2,5,6} In Western countries, such as Europe and the United States, the prevalence of HCV is as high as 30%, and is highest among people who inject drugs.⁷ In contrast, in most African countries, the prevalence of HCV is as low as 3% in Uganda⁸ and as high as 15% in Egypt.⁷

These viruses share similar modes of transmission, such as mother-to-child transmission, sharing of injecting equipment, and transfusion of unscreened blood or

blood products.^{9,10} People at high risk for HIV are also at a higher risk for other viral pathogens, including HBV and HCV. Co-infections with HBV or HCV have become increasingly common among people living with HIV. It is estimated that about one-third of people living with HIV may be co-infected with HCV and two-thirds may be co-infected with HBV, due to similar modes of transmission.⁹ These co-infections are associated with high morbidity, complications such as severe liver disease, and mortality. A diseased liver condition increases susceptibility to hepatotoxicity due to antiretroviral therapy (ART).⁹

Hepatitis co-infections have led to a heavy burden of disease in many regions of the developing world with limited resources, including Kenya, where no routine testing is available for HBV and HCV in HIV-infected patients.² Before the development and introduction of

highly active antiretroviral treatment (HAART), HIV was considered the most significant viral infection in co-infected patients, and the importance of HBV and HCV infections was underplayed.¹¹ However, since HIV infection has been successfully controlled by HAART, there has been a heightened awareness of the potentially life-threatening effects of chronic HCV or HBV infections in co-infected patients, in particular, the progression to cirrhosis and liver failure, and the development of hepatocellular carcinoma.^{2,11-13} It is possible that a person living with HIV may not know they have HBV and/or HCV infection, unless they are specifically tested for the 2 viruses.

At present, there is no vaccine for HCV. The greatest challenges to developing a vaccine include the sequence diversity between different viral groups as well as the considerable sequence heterogeneity among isolates in the N-terminal regions and the E2 glycoprotein.¹⁴ Because of this problem, it is easier to focus on secondary prevention approaches, such as screening blood products before transfusion and using single use drug injecting equipment. In contrast, a vaccine to protect contact individuals from HBV does exist, and is provided by the Kenyan government free of charge. Despite the availability of the vaccine and the fact that it is free, a number of Kenyans still have not yet received the vaccine and remain at a higher risk of contracting the disease.

In most African countries, the HIV epidemic is well documented, however, there is limited data on HBV and HCV co-infections both among HIV-infected patients and the general population.² This study was carried out among HIV-1-infected patients seeking treatment in a comprehensive care clinic, to determine the prevalence of HBV and HCV co-infections and their associated factors.

METHODS

Study Design, Setting, and Population

This cross-sectional study was carried out between May and August 2015 in Ngong Sub-County Hospital in Kajiado County, Kenya, to determine the prevalence of HBV and HCV and their associated factors among HIV-1-infected patients. The study population was comprised of HIV-infected patients attending the hospital's comprehensive care clinic, where HIV-infected individuals are reviewed and collect their antiretroviral drugs (ARVs). Patients were consecutively recruited and selected using systematic sampling method, and only those who met the inclusion criteria – 18 years of age and above, and volunteered and consented to participate in the study – were recruited. Those who were below the age of 18 years or did not consent to participate in the study were excluded from participation.

Data Collection and Laboratory Investigations

A researcher-administered questionnaire was used to collect sociodemographic information and data on factors associated with HBV and HCV co-infections. Upon completion of the

questionnaire, 5 mL of blood was aseptically collected from each participant and dispensed into EDTA vacutainer tubes to test for CD4 T-cell count, viral load, and HBV and HCV serology.

The CD4 T-cell counts were determined using BD FACSCalibre (BD, Franklin Lakes, New Jersey, USA)^{2,15} within 3 hours of sample collection, according to the manufacturer's instructions. Blood was then centrifuged for 5 minutes at 1500 revolutions per minute, and plasma was collected and dispensed in cryovials for storage at -20°C until tested. Both the hepatitis B surface antigen (HBsAg) and the hepatitis C virus IgG (anti-HCV) were determined using Maglumi 1000 (Shenzhen New Industries Biomedical Engineering Co., Ltd, Shenzhen, China), a fully automated chemiluminescence immunoassay, according to the manufacturer's instructions.¹⁶ The HBsAg kit had a sensitivity of <1 index/mL and a specificity of 100%, while the HCV IgG kit had a sensitivity of 2 U/mL and a specificity of 100%. HIV-1 viral load was determined using the Abbott m2000rt System (Abbott Molecular Inc., Des Plaines, Illinois, USA) with automated sample extraction, amplification, and detection, according to the manufacturer's instructions.¹⁷

Data Analysis

All generated data was double entered into Microsoft Excel, cleaned, and validated. The data was exported into IBM SPSS Version 20 (IBM, New York, USA) for analysis. Descriptive analysis was done for the demographic variables using frequencies and proportions. Seroprevalence for HBV and HCV was expressed as a percentage for the entire study population. Chi-square test was used to test the associations between dependent and independent variables. Odds ratios (ORs) were estimated at 95% confidence interval (CI) and the level of significance was set at *P*-value less than or equal to 0.05.

Ethical Approval

This study was approved by the Kenyatta National Hospital/University of Nairobi Ethics and Research Committee in accordance with code of ethics for biomedical research involving human subjects (reference No. P263/05/2015). The study procedure was explained in detail to all participants, and each participant signed a consent form, as an agreement to participate in this study, before answering the survey questions and providing blood samples. Consent to publish the results and patient data was obtained from the study participants in the form of a signature, after researchers explained the importance of publishing the findings of the study.

RESULTS

A total of 190 HIV-1-infected patients participated in this study. Their mean age (standard deviation [SD]) was 36.7 (10.3) years. Of the 190 participants, 150 (78.9%) were

women, while 40 (21.1%) were men. Almost two-thirds (121, 63.7%) of the participants were married. The level of education among participants was generally low, with over a tenth (24, 12.6%) having not attended school at all and about half (96, 50.5%) having attended school up to primary level. About half (51.6%) of the participants were informally employed and less than a tenth (9.5%) of the participants were formally employed. Other factors measured showed that about three-quarters (140, 73.7%) of the participants used condoms, a majority (156, 82.1%) were on ART, and almost all (181, 95.3%) were not vaccinated against HBV. All of the study participants had their CD4 and HIV-1 viral loads tested. The median (interquartile range [IQR]) viral load was 150 (150–4509) copies per mL and the median (IQR) CD4-T cell count was 469 (317–582) (Table 1).

Within the study population, the prevalence of HBV was 5.8% (n=11; 95% CI, 2.6%–8.9%) and HCV was 4.2% (n=8; 95% CI, 1.6%–7.4%). A significant difference was observed between HCV-positive and HCV-negative individuals in relation to ART (n=190; OR 0.2; 95% CI, 0.0–0.8; P=.036). For HBV, a significant difference was observed between HBV-positive and HBV-negative individuals in relation to condom usage (n=190; OR 0.3; 95% CI, 0.1–0.9; P=.039). A significant difference was also observed between HBV-positive and HBV-negative individuals in relation to median viral load. Study participants who had HIV/HBV and HIV/HCV co-infections had high median (IQR) HIV viral loads of 22570 (150–74875) and 265 (150–12867), respectively, compared to HIV viral load of 150 (150–4400) for the participants who were HIV mono-infected. Study participants who had HIV/HBV co-infection had a median (IQR) CD4 count of 350 (250–628) and HIV mono-infected participants had a median (IQR) CD4 count of 472 (321–580), while HIV/HCV co-infected participants had a median (IQR) CD4 count of 542 (400–639). Other factors, such as sex, age group, employment level, income level, education level, blood transfusion history, number of tattoos, number of sexual partners, and marital status were not significantly associated (P>0.005) with either HBV or HCV (Table 2).

DISCUSSION

In this study, the prevalence of HBsAg and HCV IgG were 5.8% and 4.2%, respectively. The occurrence of HBV was associated with condom use and HIV viral load, while HCV was associated with ART. The results of HBsAg testing in this study were consistent with HBsAg prevalence documented from previous studies carried out among HIV-infected individuals in Ethiopia (5.6%), Malawi (5.6%), and Tanzania (6.2%).^{3,11,18} The HBsAg prevalence from this study is also in agreement with a previous study conducted among HIV-infected individuals in Kenya (6%).² However, the prevalence of HIV/HBV co-infection was found lower than what has been reported in Rwanda (42.9%), Nigeria

TABLE 1. Distribution of the Participants by Socio-demographic, Behavioral, and Clinical Characteristics Among HIV-Infected Patients Attending the Comprehensive Care Clinic at Ngong Sub-County Hospital

Variable	Frequency (%)
Age, mean (SD)	36.7 (10.3)
Age	
<30 years	52 (27.4)
≥30 years	138 (72.6)
Gender	
Male	40 (21.1)
Female	150 (78.9)
Education level	
No formal education	24 (12.6)
Primary	96 (50.5)
Secondary and above	70 (36.8)
Marital status	
Married	121 (63.7)
Single	69 (36.3)
Employment	
Informal	98 (51.6)
Formal	18 (9.5)
Not working	74 (38.9)
Income (Kenya shillings)	
0	47 (24.7)
1–9999	107 (56.3)
10000–14999	18 (9.5)
≥15000	18 (9.5)
Sex partners	
0	32 (16.8)
≥1	158 (83.2)
Condom usage	
Yes	140 (73.7)
No	50 (26.4)

Continued

TABLE 1. Continued

Variable	Frequency (%)
Frequency of condom usage (n=140)	
Always	108(77.1)
Occasionally	32(22.9)
Condom breakage	
Yes	31(22.1)
No	159(77.9)
Blood transfusion	
Yes	15(7.9)
No	175(92.1)
Duration since last transfusion (n=15)	
<12 months	2(13.3)
≥12 months	13(86.7)
Tattoos	
Yes	3(1.6)
No	197(98.4)
Duration since getting the tattoo (n=3)	
<12 months	0
≥12 months	3(100)
Hepatitis B immunization	
Never	181(95.3)
Completed dose	9(4.7)
ART treatment	
Yes	156(82.1)
No	34(17.9)
CD4, median (IQR)	469(317–582)
Viral load, median (IQR)	150(150–4509)

Abbreviations: ART, antiretroviral therapy; CD4, cluster of differentiation; HBV, hepatitis B virus; HCV, hepatitis C virus; IQR, interquartile range; SD, standard deviation.

(11.9%), Ghana (11.7%), Burkina Faso (9.8%), and Myanmar (8.7%).^{4,19–22} In contrast, the prevalence of HBV in this study was higher than what has been reported in Uganda (4.1%), India (4.9%), and Australia (4.8%) among HIV-infected patients.^{23,24} In this study, use of condoms and a high HIV viral load were significantly associated with HBV

infection. High HIV viral load, as a result of high viral replication, leads to immunosuppression reducing the immune cells responsible for clearing HBV infection,^{11,15} hence the association between HBV infection and higher viral load. In addition, the association between condom usage and HBV infection can be attributed to the fact that proper condom use reduces the risk of transmission of sexually transmitted infection, such as HBV.

The prevalence of HCV in the co-infected patients in this study (4.2%) is consistent with the rates of 4.8%–5.0% reported in other studies in sub-Saharan Africa.^{11,19,23} In contrast, the prevalence of HIV/HCV was found lower than what has been reported in Kenya (10%), Ghana (18.7%), Cameroon (24.1%), Egypt (40.5%), and Australia (12.8%).^{2,4,24–26} Other examples include a study of 105 HIV-infected patients in Kathmandu, Nepal, with 13.3% HCV co-infection²⁷; a cohort of 213 HIV-infected patients in Massachusetts, USA, with 16.1% HCV co-infection²⁸; and a cohort of 639 HIV-infected individuals in Slovenia, with 7.6% HCV co-infection.²⁹ These prevalence values were higher than what is reported in this study.

However, HCV prevalence in this study was higher than that reported by previous studies in Kenya (3.7%), Uganda (3.3%), Lebanon (3.4%), Guinea-Bissau (1.7%), and Zambia (1.2%).^{8,30–33} Use of ART showed significant association with HCV infection. This could be attributed to the fact that HIV attacks immune cells leading to immunosuppression; therefore, being on ART lowers the viral load leading to immune recovery, which is responsible for HCV clearance.^{8,34}

In this study, high HIV viral loads were reported among HIV/HBV and HIV/HCV co-infected participants compared to those with HIV mono-infection. The high HIV viral loads among HIV/HBV and HIV/HCV co-infected patients may be related to increased HIV and HBV/HCV replication as well as HIV and HBV/HCV drug resistance leading to immunosuppressed state.^{15,25,34}

Study participants with HIV/HBV co-infection had low median (IQR) CD4 count compared to HIV mono-infected participants, which was incomparable with what has been reported in previous studies in South Africa (141.6) and Nigeria (121).³⁶ These controversial results may be due either to the difference in the immune status of the participants in these studies or to viral hepatitis co-infection. In individuals with both HIV and HBV, viral replication may be high, which may further contribute to the impairment of the patients' immune systems.¹¹ In addition, it is known that there is an imbalance in peripheral blood T-lymphocyte subsets and turbulence in cellular immunity in the patients with chronic HBV.³⁷ Furthermore, lamivudine-resistant mutations in HBV treatment have had adverse effects on treatment response in HIV-infected individuals co-infected with HBV, resulting in a decline in CD4 count.^{35,38,39} The median CD4 count in HIV/HCV co-infected patients was higher than in both HIV mono-infected and HIV/HBV co-

TABLE 2. Association Between Sociodemographic, Behavioral, and Clinical Characteristics With Occurrence of HBV and HCV Among HIV-Infected Patients Attending the Comprehensive Care Clinic at Ngong Sub-County Hospital

Variable	HCV Positive	HCV Negative	OR (95% CI)	P-Value	HBsAg Positive	HBsAg Negative	OR (95% CI)	P-Value
Age, mean (SD)	35.8 (8.4)	36.8 (10.4)	–	.785	38.2 (11.0)	36.6 (10.3)	–	.633
Gender								
Male	3 (7.5%)	37 (92.5%)	1	.368	4 (10.0%)	36 (90.0%)	1	.248
Female	5 (3.3%)	145 (96.7%)	0.4 (0.1–1.9)		7 (4.7%)	143 (95.3%)	0.4 (0.1–1.6)	
Age group								
<30	2 (3.8%)	50 (96.2%)	1	1	2 (3.8%)	50 (96.2%)	1	.73
≥30	6 (4.3%)	132 (95.7%)	1.1 (0.2–5.8)		9 (6.5%)	129 (93.5%)	1.7 (0.4–8.4)	
Education level								
No formal education	0 (0.0%)	24 (100.0%)	–	.998	1 (4.2%)	23 (95.8%)	1.0 (0.1–9.8)	.98
Primary	7 (7.3%)	89 (92.7%)	5.4 (0.7–45.2)	.118	7 (7.3%)	89 (92.7%)	1.8 (0.4–7.0)	.427
Secondary and above	1 (1.4%)	69 (98.6%)	1		3 (4.3%)	67 (95.7%)	1	
Marital status								
Single	5 (7.2%)	64 (92.8%)	3.1 (0.7–13.3)	.142	5 (7.2%)	64 (92.8%)	1.5 (0.4–5.1)	.532
Married	3 (2.5%)	118 (97.5%)	1		6 (5.0%)	115 (95.0%)	1	
Employment								
Informal	6 (6.1%)	92 (93.9%)	1	.998	6 (6.1%)	92 (93.9%)	1	.449
Formal	0 (0.0%)	18 (100.0%)	–	.305	2 (11.1%)	16 (88.9%)	1.9 (0.4–10.3)	.549
Not working	2 (2.7%)	72 (97.3%)	0.4 (0.1–2.2)		3 (4.1%)	71 (95.9%)	0.6 (0.2–2.7)	
Sex partner								
0	2 (6.3%)	30 (93.8%)	1	.624	3 (9.4%)	29 (90.6%)	1	.4
≥1	6 (3.8%)	152 (96.2%)	0.6 (0.1–3.1)		8 (5.1%)	150 (94.9%)	0.5 (0.1–2.1)	
Income (Kenya shillings)								
0	0 (0.0%)	47 (100.0%)	–	.998	1 (2.1%)	46 (97.9%)	0.4 (0.0–6.2)	.49
1–9999	6 (5.6%)	101 (94.4%)	1.0 (0.1–8.9)	.993	8 (7.5%)	99 (92.5%)	1.4 (0.2–11.7)	.771
10000–14999	1 (5.6%)	17 (94.4%)	1.0 (0.1–17.3)	1	1 (5.6%)	17 (94.4%)	1.0 (0.1–17.3)	1
≥15000	1 (5.6%)	17 (94.4%)	1		1 (5.6%)	17 (94.4%)	1	
Condom usage								
Yes	6 (4.3%)	134 (95.7%)	1.1 (0.2–5.5)	1	5 (3.6%)	135 (96.4%)	0.3 (0.1–0.9)	.039
No	2 (4.0%)	48 (96.0%)	1		6 (12.0%)	44 (88.0%)	1	
Condom breakage								
Yes	2 (6.5%)	29 (93.5%)	1.8 (0.3–9.1)	.619	2 (6.5%)	29 (93.5%)	1.1 (0.2–5.6)	1
No	6 (3.8%)	153 (96.2%)	1		9 (5.7%)	150 (94.3%)	1	
Transfusion								
Yes	0 (0.0%)	15 (100.0%)	–	1	0 (0.0%)	15 (100.0%)	–	1
No	8 (4.6%)	167 (95.4%)			11 (6.3%)	164 (93.7%)		

Continued

TABLE 2. Continued

Variable	HCV Positive	HCV Negative	OR (95% CI)	P-Value	HBsAg Positive	HBsAg Negative	OR (95% CI)	P-Value
Tattoos								
Yes	1 (33.3%)	2 (66.7%)	12.9 (1.0-159.2)	.122	0 (0.0%)	3 (100.0%)	-	1
No	7 (3.7%)	180 (96.3%)	1		11 (5.9%)	176 (94.1%)		
ARV treatment								
Yes	4 (2.6%)	152 (97.4%)	0.2 (0.0-0.8)	.036	7 (4.5%)	149 (95.5%)	0.4 (0.1-1.3)	.111
No	4 (11.8%)	30 (88.2%)	1		4 (11.8%)	30 (88.2%)	1	
Hepatitis B immunization								
Finished	-	-	-	-	0 (0.0%)	9 (100.0%)	-	1
Never					11 (6.1%)	170 (93.9%)		
CD4, median (IQR)	542 (400-639)	465 (312-578)	-	.423	350 (250-628)	472 (321-580)	-	.431
Viral load, median (IQR)	265 (150-12867)	150 (150-4400)	-	.399	25570 (150-74875)	150 (150-3078)	-	.002

Abbreviations: ART, antiretroviral therapy; CD4, cluster of differentiation; HBV, hepatitis B virus; HCV, hepatitis C virus; IQR, interquartile range; SD, standard deviation.

infected study participants. Why the median CD4 count in HIV/HCV co-infected study participants was higher than those with HIV/HBV co-infection was unclear.

The predominance of women in this study could be a reflection of higher burden of HIV in women in Kenya as well as the observation that men find it harder to reveal their HIV status, which leads to poorer health-seeking behaviour for HIV care services. However, the role of gender in the disparity of HBV and HCV burden is not fully known.

Studies show that co-infection rates of HBV and HCV in HIV-infected individuals vary globally depending on type of exposure, risk group, geographical region, sensitivity and specificity of the test kits, and difference in sample sizes. This study was limited in several aspects; mainly the anti-HCV- and HBsAg-positive samples were not confirmed by polymerase chain reaction to rule out false-positive results or repeated thawing and freezing of the sample, which may affect the results. In addition, because this was a cross-sectional study, we could not determine or confirm when the study participants acquired HBV and HCV infections and, therefore, could not establish a temporal relationship between risk factor and outcome. Furthermore, the study results relied on self-reported data, which meant that as participants retrospectively reported their lifetime behaviours, inaccuracies may have been introduced through recall bias.

CONCLUSIONS

Our study showed a high prevalence of HBV (5.8%) and HCV (4.2%) co-infection among HIV-1-infected patients.

The results of this study also found that factors associated with HIV/HBV co-infection were condom use and HIV viral load, and HIV/HCV co-infection was associated with ART. Based on the findings of this study, the authors suggest that HIV-infected patients should be routinely tested for HBV and HCV. In addition, health education should be provided on an ongoing basis to HIV-infected patients on issues such as the importance of HIV treatment, use of protection while having sex, and monitoring of HIV infection. Furthermore, we call on policy makers and health providers to put in place preventive and control measures that include public education on HBV and HCV infections.

Acknowledgements: The authors wish to thank and acknowledge Ngong Sub-County Hospital for permission to carry out the research and the study participants whose participation made it possible for this outcome.

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Peer Reviewed**Competing Interests:** None declared.**Received:** 26 Oct 2016; **Accepted:** 25 Jun 2017**Cite this article as:** Ndegwa Maina D, Kimanga Nyerere A, Wambui Gicho R, Maina Mwangi J, Wekesa Lihana R. Prevalence and Factors Associated With Hepatitis B and C Co-Infection Among HIV-1-Infected Patients in Kenya. *East African Health Res J.* 2017;1(2):73-79. <https://doi.org/10.24248/EAHRJ-D-16-00334>© Maina et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are properly cited. To view a copy of the license, visit <http://creativecommons.org/licenses/by/3.0/>. When linking to this article, please use the following permanent link: <https://doi.org/10.24248/EAHRJ-D-16-00334>

Seroprevalence and Knowledge of Hepatitis B Virus Infection Among Laboratory Workers at Kilimanjaro Christian Medical Centre in Moshi, Tanzania

Richard E Machange,^{a,b} Dominic Mosha,^c Jeremia J Pyuza,^b Balthazar B Nyombi,^b Elichilia R Shao^{a,d,e}

^aDepartment of Internal Medicine, College of Medicine, Kilimanjaro Christian Medical University College, Moshi, Tanzania; ^bDepartment of Clinical Laboratory, College of Medicine, Kilimanjaro Christian Medical University College, Moshi, Tanzania; ^cDepartment of Community Medicine, College of Medicine, Kilimanjaro Christian Medical University College, Moshi, Tanzania; ^dBetter Human Health Foundation, Moshi, Tanzania; ^eImagedoctors International, Arusha, Tanzania.
Correspondence to Elichilia R Shao (Elichilia2004@yahoo.co.uk).

ABSTRACT

Background: Hepatitis B Virus (HBV) is transmitted through blood, infected body fluids, and unsterile needles and surgical equipment. We first determined the current seroprevalence of HBV and vaccination coverage, then assessed knowledge on risk factors for hepatitis B virus infection among medical laboratory workers at Kilimanjaro Christian Medical Centre (KCMC) in Moshi, Tanzania.

Methods: A cross-sectional study was conducted from January to June 2014, involving health care workers (HCWs) from KCMC. Eligibility for participation in the study was determined by length on employment, provision of consent, and willingness to complete a questionnaire. Recruitment was non-randomised; a simple and consecutive sampling of 85 eligible HCWs was conducted at the hospital during study period. Structured questionnaires were self-administered and consenting participants allowed blood samples to be tested for HBV infection. Blood (4 mL) was obtained by venepuncture from all participants using sterilised disposable 5 mL syringes and 20 gauge needles. All collected blood was tested for HBsAg using enzyme linked immunosorbent assay according to manufacturer's guidelines. A cut-off point of 10% ($P > 0.1$) was used to select variables to be included in the further analysis.

Results: Out of the 76 HCWs eligible to participate in the study, only 8 (10.5%) were vaccinated against HBV. Of the 68 unvaccinated laboratory workers, 9 (11.8%) tested positive for HBsAg. Knowledge about HBV infection and its associated risks was high among medical personnel – where 36.4% scored above 65% – compared with non-medical personnel, none of whom scored as high as 65%.

Conclusions: Seroprevalence of HBsAg among laboratory workers at KCMC was 11.8%. Low knowledge of risks for HBV infection placed HCWs at great risk of occupational exposure. Low vaccination coverage among HCWs increases the risk of acquiring HBV infection following occupational exposure.

BACKGROUND

Hepatitis B virus (HBV) infection is among the viral diseases responsible for serious liver diseases, such as cirrhosis and hepatocellular cancer. Viral hepatitis is a serious public health problem affecting billions of people. Globally, approximately 240 million people are chronically infected with HBV.^{1,2} The prevalence of HBV has been reported to be higher in East Asia and sub-Saharan Africa.^{1,2} In countries with high HBV prevalence, most HBV infections happen during childhood. Of the children who are not infected, many remain at risk for contracting the disease as adults, if they are not

vaccinated.³ For health care workers (HCWs), the risk of contracting HBV is much higher than the general population, due to the nature of the work they do daily.⁴ A study in Niger demonstrated that HCWs have 4 times the risk – compared with the general population – of becoming infected with HBV through direct contact, including needle sticks with infectious materials like HBV-infected blood, blood products, or contaminated fluids.⁵

In developing countries, 50% of HBV infection among HCWs has been attributed to professional hazard.⁶ Studies conducted in Pakistan and Nigeria indicate that poor knowledge, leading to inadequate precautions against blood-borne infection; lack of adequate training

on infection prevention control of hospital-acquired infections; and low coverage of vaccination against HBV, among others, contribute to the high prevalence of HBV among HCWs.^{7,8}

Through vaccination, HBV infection is a preventable disease. Despite the availability of HBV vaccines, HCW vaccination rates are not sufficient and significant numbers of HCWs are unvaccinated or undervaccinated. The latter refers to HCWs not receiving all three of the required doses,⁹ or receiving all three doses but, because of potential poor immune response, requiring an antibody titre to see if they mounted enough antibodies for protection.^{10,11} Poor immune response to vaccination can be attributed to old age, diabetes mellitus, other viral infections, and low body mass index, all of which contribute to weak immunogenicity. Because of these multiple factors, the World Health Organization (WHO) recommends compulsory vaccination for HCWs that includes additional monitoring of their immune response after vaccination.⁹⁻¹³

In Tanzania, the prevalence of HBV infection among HCWs in tertiary hospital in Mwanza was found to be 7.0%.¹⁴ This prevalence is similar to a study of HBV infection among blood donors in Dar es Salaam, Tanzania, which found 8.8% prevalence in that study population.¹⁴ Low knowledge of the risks for HBV infection place medical laboratory workers at risk for and low vaccination coverage among HCWs increases the risk of acquiring HBV infection following occupational exposure. The aim of our study is to provide information on the seroprevalence and vaccination coverage of and risk factors for HBV infection among HCWs at the Kilimanjaro Christian Medical Centre (KCMC) referral hospital in Moshi, Tanzania.

METHODS

Study Site and Design

KCMC is a tertiary referral and teaching hospital in the Kilimanjaro region of Tanzania. It is 1 of the 4 zonal consultant hospitals in the country, and is the referral hospital for over 15 million people in Northern Tanzania.¹⁵ The centre's laboratory is vast complex with multiple units that specialise in different clinical areas and employs 85 people. A cross-sectional study was conducted from January to June 2014, involving HCWs who work in the centre. Study participants were recruited from clinical and biotechnology laboratories, which provide services to all patients coming to the centre.

Methods and Tools

The study conducted questionnaires with and collected biological samples from HCWs who met the study's inclusion criteria. The principal investigator completed the structured questionnaire for each HCW included in the study. The questionnaire had 4 sections: demographic and academic characteristics, knowledge of the risk factors for HBV infection and vaccines, history of accidental exposure to blood and

its products, and the perception of HBV vaccines and vaccination status. A detailed history was obtained from each HCW about their HBV vaccination history and any occupational risks, such as exposure to infected blood through a splash, needle stick, or cut/wound. Other invasive procedures, such as intravenous therapy, intramuscular or subcutaneous injections, blood transfusions, and surgery were recorded. Identity (ID) numbers were written or placed on each participant's questionnaire and blood container to ensure the questionnaire and blood sample were matched to the same person.

Study Participants and Sampling

To be eligible to participate in the study, HCWs must have been employed for more than 3 months, able and willing to provide consent, and willing to complete the self-administered questionnaire. Recruitment was non-randomised; a simple and consecutive sampling of eligible HCWs was conducted at the hospital during study period. The KCMC laboratory had total number of 85 workers, of which 3 did not consent, 3 had been employed for less than 3 months, and 3 did not complete the questionnaire, and were therefore excluded from the final study population of 76 HCWs. Every consenting participant was given a hard copy of the questionnaire to complete at their will and convenience. Participants were given a unique ID number which was used in place of their names, allowing the questionnaires and blood samples to be paired and submitted anonymously to the researchers.

Blood Collection

Blood (4 mL) samples were obtained by venepuncture from all consenting participants using sterilised disposable 5 mL syringes and 20 gauge needles. A red top bottle was labelled with the study participant's specific study ID number. An hour after the blood samples were drawn, they were processed at the KCMC clinical laboratory.

Sample Processing

All collected blood samples were tested for the hepatitis B antigen (HBsAg) using enzyme linked immunosorbent assay (ELISA) according to the manufacturer's guidelines. Serums/plasma were added to antigens or antibodies fixed to a solid surface microplate, incubated, and then washed using Thermo Scientific Wellwash Versa. The samples were then taken to Thermo Scientific Multiskan FC microplate photometer reader, with Skanlt Software, to be scanned and processed. To ensure this process provided correct results, rapid tests for HBsAg were used to re-test any positive test for HBsAg. All the methods used were in accordance with government laboratory standards. In the context of the current study, a person was considered to be fully vaccinated after receiving a minimum of intramuscular injection of 1 mL of DNA-recombinant vaccine scheduled at 0, 1, and 6 months, thus completing the minimum of the required

3-vaccine series.¹⁶ Participants who did not receive the 3 doses were considered to be inadequately vaccinated or undervaccinated, while those who did not receive the single dose were considered unvaccinated.

Statistical Analysis

Data were captured in Microsoft Excel and transferred to Stata version 13.1 (StataCorp, College Station, Texas, USA) for analysis. Descriptive analysis was performed whereby numeric variables were summarised using measures of central tendency and the corresponding measures of dispersion. Categorical variables were summarised using frequency and percentages. Chi-square (X^2) test was used to compare the prevalence of hepatitis B infection by different participant characteristics.

Multivariable logistic regression (odds ratio and the corresponding 95% confidence interval) was used to determine factors associated with hepatitis B infection among HCWs at KCMC. A cut-off point of 10% ($P>0.1$) was used to select variables to be included in the further analysis. After controlling for potential confounders, variables with $P<0.05$ were considered to be statistically associated with hepatitis B infection.

Ethical Approval

The study was conducted following ethical approval from the Kilimanjaro Christian Medical University College Review Board and approval from the medical director of KCMC. Written informed consent was obtained from all participants.

RESULTS

Among the 76 study participants, 40 (52.6%) were men and 36 (47.4%) were women ages 20–56 years with a duration of employment ranging from 3 months to 29 years. Health histories of the group revealed that 8 (10.5%) had a history of vaccination, 7 (9.2%) had a history of blood transfusion, 5 (6.6%) had history of mucocutaneous exposure to HBV, and 3 (3.9%) had history of surgery. Among the 9 (11.8%) HCWs who tested positive for HBsAg from unvaccinated group, 6 (66.7%) were medical personnel, 2 (20.0%) were administrators who had previous history of surgery and blood transfusion, and 1 was an information technology (IT) specialist. The characteristics and distribution of participants with HBV positive and negative results are summarised in Table 1. The frequency of hepatitis B infection among medical personnel was 9.1%, while among non-medical personnel was 30%. In this study, only 7 HCWs had been vaccinated against HBV – 4 received full vaccination coverage, 2 received only 2 shots, and the last received only 1 shot of the vaccine. None of the workers had their antibody status checked after finishing the full 3 shots of vaccine, which is WHO protocol.

The questionnaire given to participants assessed their knowledge of HBV infection and vaccination, scores were

grouped into low (<50%), moderate (50%–74%), and high ($\geq 75\%$) categories (Table 1).¹⁷ Knowledge about HBV infection and vaccination was higher among men compared to women. Knowledge was higher among medical personnel and very low among non-medical personnel – about 5 (50%) of all non-medical personnel had low knowledge about HBV infection and vaccination, as compared to 8 (12%) of medical personnel (Table 1).

On assessing the risks, out of the 66 medical personnel studied, 6 (9.1%) were positive for HBsAg; and among those 6, 5 (83%) had positive history of blood transfusion and only 1 had had surgery. For non-medical personnel, 3 were positive for HBsAg and 2 (67%) had a positive history of blood transfusion.

DISCUSSION

The current study revealed a significant burden of HBV infection among HCWs at the KCMC referral and teaching hospital in Moshi, Tanzania. The overall seroprevalence of HBsAg among HCWs at KCMC was 11.9%. This prevalence is higher than a similar study conducted in Tanzania's Lake Zone, which found 7% prevalence among HCWs – doctors, medical laboratory workers, nurses, IT experts, among others.¹⁴ In the neighbouring country of Uganda, a similar study was conducted among HCWs, which revealed a prevalence of 8.1%.^{4,14} Among the 6 medical attendants who participated in the KCMC study, 3 (50%) were infected with HBV, most of whom scored very low on hepatitis B knowledge on the questionnaire. This may be explained by their higher level of exposure to infected material, such as used needles and soaked gauze, which increased their risk of infection; and by their low knowledge of the disease and its risk factors, which may have affected their behaviour towards using protective gear to reduce their risk.

In this study, potential risk exposures among HCWs, as a whole, might be among the predisposition to occupational accidents, as reported in other studies.^{16–19} However, of the 5 IT specialists in our study, 2 were infected with HBV. This could be explained by the fact that they receive and enter blood sample information into the laboratory information system, which exposes them to the blood products. Other possible explanations for their level of infection is their exposure to HBV through blood transfusion and surgery or their poor knowledge about HBV infection and vaccination. Although the study had limited information about their immune status information prior to employment, occupational risk factors for contracting HBV in the laboratory environment cannot be completely excluded. The incidence of occupational exposure is different from one facility to another depending on the quality of facility and knowledge of the workers as well as the prevalence of HBV in the general population.¹⁸

The current study found that a quarter of the study participants were above 40 years of age. Most of the HCWs who

TABLE 1. Positivity and Risk Factors of Occupational Exposure in Health Care Workers at KCMC, Moshi Tanzania

Characteristics	Number	Positive Hepatitis B Surface Antigen n (%)	Reported 'Yes' for Below Potential Exposures of Infection					Knowledge on HBV Infection and Vaccination		
			Blood Transfusion n (%)	IV/IM n (%)	Operation n (%)	Muco-cutaneous Exposure n (%)	Vaccinated n (%)	Low n (%)	Moderate n (%)	High n (%)
Sex										
Male	40	4 (10.0)	3 (7.5)	5 (12.5)	1 (2.5)	4 (10.0)	5 (12.5)	6 (15.0)	15 (37.5)	19 (47.5)
Female	36	5 (13.9)	4 (11.1)	8 (22.2)	0 (0.0)	3 (8.3)	2 (5.6)	7 (19.4)	24 (66.7)	5 (13.9)
Age (Years)										
<40	49	4 (8.2)	4 (8.2)	1 (2.0)	3 (6.1)	6 (12.2)	3 (6.1)	9 (18.4)	27 (55.1)	13 (26.5)
≥40	27	5 (18.5)	3 (11.1)	12 (44.4)	0 (0.0)	1 (3.7)	4 (14.8)	4 (14.8)	12 (44.4)	11 (40.7)
Cadre										
Medical personnel	66	6 (9.1)	5 (7.6)	13 (19.7)	1 (1.5)	7 (10.6)	7 (10.6)	8 (12.1)	34 (51.5)	24 (36.4)
Non-medical personnel	10	3 (30.0)	2 (20.0)	0 (0.0)	2 (20.0)	0 (0.0)	0 (0.0)	5 (50.0)	5 (50.0)	0 (0.0)
Duration at Work (Years)										
≤5	44	4 (9.1)	4 (9.1)	2 (4.5)	2 (4.5)	2 (4.5)	2 (4.5)	9 (20.4)	25 (56.8)	10 (22.7)
>5	32	5 (15.6)	3 (9.4)	11 (34.4)	1 (3.1)	2 (6.3)	5 (15.6)	4 (12.5)	14 (43.7)	14 (43.7)

Abbreviations: KCMC, Kilimanjaro Christian Medical Centre; IV, intravenous puncture; IM, Intramuscular puncture.
 Notes: Knowledge was measured by asking 20 standardized questions. Each question carries 5 marks: Therefore 5X20=100%.
 Grading knowledge basing on the obtained scores by the study participants
 (1) Low knowledge ≤40%
 (2) Moderate knowledge 41–65%
 (3) High >65%

were infected were from this group. One explanation for this may be that HCWs have greater exposure to HBV during their lifetime; hence, prevalence may increase with age. Another explanation, which is consistent with other studies, is that long occupational exposure – number of years of employment – increase chances for HBV acquisition.^{14,20}

To date, Tanzania has not performed a controlled and monitored nationwide vaccination campaign among high-risk groups, such as HCWs, as recommended by WHO.¹⁶ Very few HCWs have been vaccinated because vaccinations were only given as a donation or as a precondition for clinical research work.^{4,21} This study assessed vaccination coverage among HCWs and found it to be very low, which is also seen in other developing countries. The KCMC study's level of vaccination coverage among HCWs was 8.8%. In contrast, vaccination coverage of medical students in Cameroon was

18%; however, only 10% of the fully vaccinated students had their antibody levels checked.^{20,21} In this study, of the few who had been vaccinated, some did not finish the full vaccine series; and none of the HCWs who did finish the full dosage series, had an antibody titre to confirm immunity, which is recommended by WHO. A study in Karachi reported a high vaccination coverage of 81.8%,²² however, the study population's level of immunity was not confirmed with an HBsAg titre.

The KCMC study assessed the level of knowledge on HBV infection and vaccination among HCWs, which was found to be generally very low. Good knowledge about HBV vaccination and infection may reduce infection rate. A study conducted in the Sudan showed that some medical laboratory workers did not know that HBV can lead to liver cancer, they only knew about the relationship between cirrhosis

and liver cancer.^{23–25} This suggests more training is needed for HCWs on HBV infection and vaccination.

In the KCMC study, knowledge was higher among medical personnel compared to non-medical personnel. This could be attributed to difference in the type of education each group received. However, some cases of low knowledge among medical personnel have been reported; for example, literature from Iran showed poor knowledge among medical doctors about HBV infection occurring through vaginal route.^{26,27}

We assessed different factors, such as surgery and blood transfusion, that contributed to HBV infection. Out of the 6 medical and 3 non-medical personnel who tested positive for HBV, 5 and 2, respectively, had history of blood transfusion. These factors likely contributed to the high prevalence among these particular participants. A study conducted in Gabon showed the contribution of blood transfusion and surgical interventions towards increasing seroprevalence of HBV.²⁸ Our study found no difference in positivity between participants with many years of work experience compared with those workers with only a few years.²⁹

Limitations

Due to small sample size of our study, which focused only to personnel within a laboratory setting, our findings are not generalizable to HCWs in other settings. All information about HBV vaccination status was obtained through interviews without laboratory confirmation of an antibody titre, which may have introduced the possibility of interviewee bias.

CONCLUSIONS

This study showed high prevalence of HBV and low coverage of HBV vaccination among laboratory workers. Low knowledge of risks for HBV infection may have placed these workers at great risk of occupational exposure. Low vaccination coverage among HCWs increases the risk of acquiring HBV infection following occupational exposure. The aim of this study was to provide a further evidence of a major problem that needs immediately attention and action to be taken. Tanzanian government has done great work for HBV vaccination among children under 5; we suggest the same calibre of effort must be considered for workers in all specialties.

Acknowledgement: Authors acknowledge all workers at KCMC clinical laboratory and biotechnology for participating in this research. We are grateful for permission from the KCMC Executive Director as well as the laboratory manager of biotechnology for permission to conduct this research at their centres. The KCMC has always been keen and generous in fostering research in order to offer Tanzanians a better quality of life. We also wish to extend our thanks and appreciation to Kilimanjaro Christian Medical University College for its support during this study. Votes of thanks also go to the Community Health Department Director General of the Health Affairs of the Kilimanjaro Christian

Medical University College. We are indebted to Dr. Nelson Urima for his assistance.

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Peer Reviewed**Competing Interests:** None declared.**Received:** 1 Nov 2016; **Accepted:** 24 Feb 2017**Cite this article as:** Machange RE, Masha D, Pyuza JJ, Nyombi BB, Shao ER. Seroprevalence and Knowledge of Hepatitis B Virus Infection Among Laboratory Workers at Kilimanjaro Christian Medical Centre in Moshi, Tanzania. *East African Health Res J.* 2017;1(2):80-85. <https://doi.org/10.24248/EHRJ-D-16-00362>

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Factors Associated With *Toxoplasma gondii* IgG and IgM Antibodies, and Placental Histopathological Changes Among Women With Spontaneous Abortion in Mwanza City, Tanzania

Illuminata Machumi,^a Mariam M Mirambo,^b Deodatus Ruganuzi,^c Peter Rambau,^d Anthony N Massinde,^a Albert Kihunrwa,^a Stephen E Mshana,^b Domenica Morona^c

^aDepartment of Obstetrics and Gynaecology, Weill Bugando School of Medicine, Mwanza, Tanzania; ^bDepartment of Microbiology and Immunology, Weill Bugando School of Medicine, Mwanza, Tanzania; ^cDepartment of Medical Parasitology and Entomology, Weill Bugando School of Medicine, Mwanza, Tanzania; ^dDepartment of Pathology, Weill Bugando School of Medicine, Mwanza, Tanzania.
Correspondence to Domenica Morona (dmorona@gmail.com)

ABSTRACT

Background: *Toxoplasma gondii* infection in early pregnancy has been associated with significant adverse pregnancy outcomes. Despite being common in the city of Mwanza, its association with spontaneous abortion has never been studied. Here, we report the IgG and IgM seropositivity and histopathological changes of toxoplasmosis among women with spontaneous abortion.

Methods: A total of 260 women with spontaneous abortion were enrolled between November 2015 and April 2016 from 4 hospitals in Mwanza city. Specific *T. gondii* IgG and IgM antibodies were detected from sera by indirect enzyme-linked immunosorbent assay (ELISA) while the conceptus tissues were stained with haematoxylin and eosin to demonstrate histopathological changes. Data were analysed by using Stata version 13.

Results: The mean age of the enrolled women was 26±5.9 years. The seropositivity of IgG and IgM antibodies were 144/260 (55.4%; 95% confidence interval [CI], 49–61) and 6/260 (2.3%; 95% CI, 3–8), respectively. IgG seropositivity was significantly high among women in the first trimester (59.1% vs. 43.5%; $P=0.03$). Only low gestation age (odds ratio [OR] 1.11; 95% CI, 1.02–1.20; $P=0.02$) and keeping a cat (OR 11.80; 95% CI, 1.32–10.5; $P=0.03$) independently predicted IgG and IgM seropositivity, respectively. Presence of inflammation (OR 1.95; 95% CI, 1.05–3.64; $P=0.03$), calcification (OR 3.28; 95% CI, 1.01–10.63; $P=0.04$), necrosis (OR 2.86; 95% CI, 1.39–5.89; $P=0.04$), and lymphocyte infiltrations (OR 2.24; 95% CI, 1.17–4.24; $P=0.01$) were significantly associated with *T. gondii* IgG seropositivity.

Conclusions: Almost half of women with spontaneous abortion in the city of Mwanza have specific *T. gondii* IgG antibodies. Placental histopathological changes suggestive of toxoplasmosis were significantly found among IgG seropositive women. This calls for the need to screen these women during antenatal visits in order to institute appropriate measures, such as treatment and counselling, to prevent complications associated *T. gondii* infection.

BACKGROUND

Toxoplasmosis is a common infection caused by a coccidian intracellular protozoan parasite, *Toxoplasma gondii*, which occurs in domestic animals and humans throughout the world. It is a public health concern by reason of its neurological manifestations among human immunodeficiency virus/acquired immunodeficiency syndrome (HIV/AIDS) patients and the potential association with adverse pregnancy outcomes.^{1–4} The

infection is mainly acquired through ingestion of undercooked or raw meat containing viable cysts, ingestion of food and water contaminated with oocysts shed by cats, or congenitally during pregnancy.^{5–7} The course of the primary infection is often subclinical, with a majority of the infected individuals remaining asymptomatic; few patients may present mild symptoms.^{8,9} Primary infections during pregnancy are often asymptomatic but may result into foetal complications like spontaneous abortions, stillbirths, severe congenital

malformations, and central nervous system symptoms in apparently normal infants.^{10–13} There is a geographical variation of the epidemiology of *T. gondii* infection. One-third of the world's population is estimated to be infected with *T. gondii*.^{14,15} About 0.01% to 0.1% of infants in developed countries are affected by congenital toxoplasmosis.¹⁶ Primary infection during the third trimester carries a higher risk of congenital infection than infection in the first and second trimesters.^{16–18} However, severe foetal sequels occur when the disease is acquired in the first trimester.^{8,19} Though data from African countries are still scarce, a few studies carried out in Tanzania have documented the magnitude of toxoplasmosis,^{20–23} however, none of these studies focused on women with spontaneous abortion. A high prevalence of toxoplasmosis among women with spontaneous abortions has been reported in Egypt and Mexico.^{24,25} Given the fact the *T. gondii* seropositivity among pregnant women is high in Mwanza²⁰ there is a paramount need to investigate its association with poor pregnancy outcomes. The current study was undertaken in Mwanza to investigate the role of *T. gondii* infection as a potential cause of spontaneous abortion. The data collected may inform policy makers and prompt them to consider the need for a policy of screening and treatment of this infection during pregnancy to reduce possible associated complications.

MATERIALS AND METHODS

Study Design and Study Area

A cross-sectional hospital-based study was conducted between November 2015 and April 2016 involving 4 health facilities in the city of Mwanza, Tanzania. The 4 facilities included the Bugando Medical Centre (BMC), Sekou Toure regional hospital, Nyamagana district hospital, and Buzuruga health centre. These sites were purposively selected because they serve a large population of the city and provide obstetrics and gynaecological services for women with spontaneous abortion.

Study Population and Inclusion and Exclusion Criteria

The study included all women diagnosed with spontaneous abortion in their first and second trimester of the pregnancy attending obstetrics and gynaecology clinics and emergency departments at the 4 selected sites. Women who were unsure about the dates of their last normal menstrual period and those in critical condition were excluded from the study.

Sample Size Estimation and Sampling Techniques

The sample size was estimated by the Kish Leslie formula,²⁶ using the prevalence of 12.8% from Kistiah et al.²⁷ The minimum sample size calculated was 174, however a total of 260 women were enrolled. A serial sampling technique was used to enrol participants until the desired sample size was reached.

Data Collection

Sociodemographic and medical/obstetric information was collected by a direct assessment of the study participants and pre-tested structured questionnaires.

Sample Collection Procedure and Laboratory Investigations

During the collection of tissue samples, a checklist was provided to exclude observable signs of induced abortion, such as lacerations, cervical bruises, and foreign bodies during evacuation. A small sample of conceptus was collected and placed into 10% neutral buffered formalin for fixation. The tissues were subsequently processed and stained by haematoxylin and eosin as previously described.²⁸ Slides were read by an experienced pathologist to detect the presence of necrosis, calcifications, plasma cells, and different forms of inflammation and to identify tachyzoites and bradyzoites.

For serological diagnosis, about 5 mL of venous blood was collected aseptically using plain vacutainer tubes (Becton, Dickinson and Company, Nairobi, Kenya). The samples were then taken to the Catholic University of Health and Allied Sciences (CUHAS) multipurpose laboratory where the serum was separated by centrifugation at 3,000 rpm for 5 minutes. The sera were kept at -40° C until processing. The detection of specific *Toxoplasma* IgM and IgG antibodies was done by commercial indirect enzyme-linked immunosorbent assay (ELISA) (PishtazTeb Diagnostics, Teheran, Iran). The IgM ELISA assay used IgM capture principle. All procedures followed manufacturer instructions. A standard curve for IgG antibody detection was obtained by calibrating the standards 1 to 5 with concentration of 0, 10, 50, 100, and 200 IU/mL using ChemWell 2910 Automated EIA (Awareness Technology, Inc., Palm City, Florida, USA) as per manufacturer's instructions.

Data Analysis

Data were entered into Microsoft Office Excel 2013. Coding and analysis were carried out using Stata version 13 (StataCorp, College Station, Texas, USA). Continuous variables, such as age, gestation age, gravidity, and antibody titres were summarized as median with interquartile range (IQR) or means with standard deviation. Categorical variables such as marital status, residence, education level, occupation, history of miscarriage, history of stillbirths, HIV status, keeping a cat, drinking unboiled water, or consuming pork, chicken, mutton, lamb, or beef were summarized as proportions. For the histopathological results, data were analysed using the Pearson's Chi-square test to observe the statistical differences of proportions in the various groups. In addition, the Wilcoxon Ranksum (Mann Whitney) test was used to compare differences on the medians between the groups. Univariate and multivariate logistic regression models were used to determine the predictors of *T. gondii* infection. Predictors with *P*-value of less than 0.2 were

subjected into multivariate logistic regression analysis and their ORs and 95% CIs were noted. Predictors with *P*-values of less than 0.05 were considered statistically significant.

Ethics approval and consent to participate

Ethical approval was obtained from the joint CUHAS/BMC research ethics and review committee with ethical clearance number CREC/103/2015. Written informed consent was obtained from each participant prior to enrolment in the study. For participants aged below 18 years, consent was given by the parents/guardians who accompanied them.

RESULTS

Sociodemographic Characteristics of the 260 Women Enrolled in the Study

The mean age of enrolled women was 26±5.9 years. The majority (198, 76.2%) were in the first trimester, and 160 (61.5%) resided in rural areas. A total of 147 (56.5%), 86 (33.1%), and 27 (10.4%) had either no formal/primary, secondary, or tertiary education, respectively. The median gestation age of the enrolled women was 11 (IQR 9–13) weeks. A total of 114 (43.8%) of the enrolled women were housewives, while 58 (19.2%) and 88 (33.8%) were peasants or employed, respectively. The majority of women 185 (71.1%) were either primipara—a woman who is giving birth for the first time—or had at least 1 previous birth, as shown in Table 1.

Seropositivity of Specific *T. gondii* Antibodies Among Women with Spontaneous Abortion

The seropositivity of IgG antibodies was 144/260 (55.4%; 95% CI, 49–61). Of the 198 women who were in the first trimester, 117 (59.09%) were IgG seropositive compared to only 27/62 (43.5%) of those in second trimester (*P*=.03). Regarding IgM seropositivity, the seropositivity was 6/260 (2.3%; 95% CI, 3–8).

Factors Associated with Specific *T. gondii* Antibodies Among Women with Spontaneous Abortion

The median gestation age of IgG seropositive women was significantly lower than the median gestation age of IgG seronegative women (11 IQR 9–13 vs. 12 IQR 10–14; *P*=.02). On univariate logistic regression analysis, the decrease in gestation age (OR 1.09; 95% CI, 1.10–1.19; *P*<.03) was significantly associated with IgG seropositivity (Figure 1). There was no significant difference between the mean age of IgG seropositive women and that of IgG seronegative women (26±5.9 vs. 25±5.8; *P*=.17).

Out of 4 participants with HIV-positive status, 3 (75%) were IgG seropositive compared to 105 (55.3%) and 36 (54.6%) of those with HIV-negative status and those with unknown status, respectively (*P*>.05). Other factors

TABLE 1. Sociodemographic Characteristics of the 260 Women With Spontaneous Abortion Enrolled in the Study

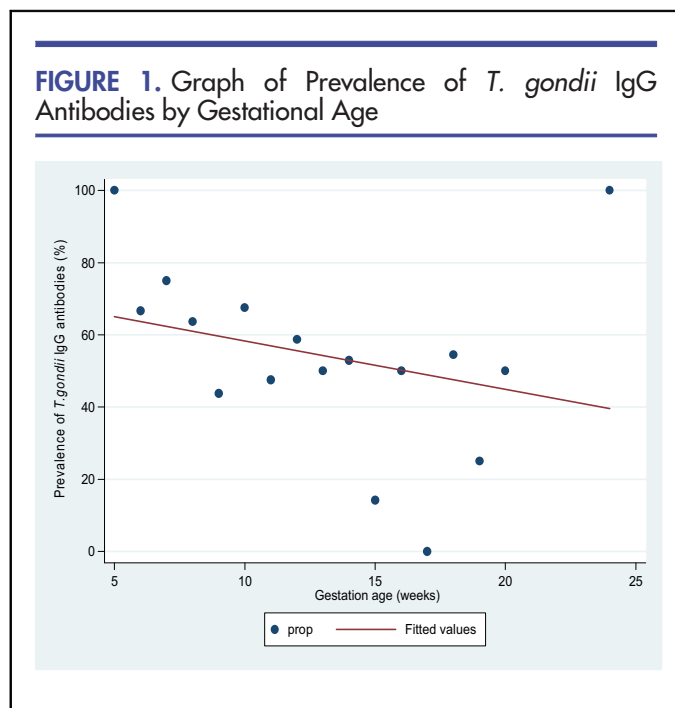
Characteristics	n (%)
Age*	26±5.9
Gestation age*	11 (IQR 9–13)
First trimester	198 (76.2)
Second trimester	62 (23.8)
Education	
No formal/primary	147 (56.5)
Secondary	86 (33.1)
Tertiary	27 (10.4)
Residence	
Rural	160 (61.5)
Urban	100 (38.5)
Occupation	
Housewife	114 (43.8)
Peasant	58 (19.2)
Employed	88 (33.8)
Parity	
Nullipara/0 children	75 (28.9)
≥1 children	185 (71.1)

* Mean age and median gestation age of the study participants. Abbreviation: IQR, interquartile range.

investigated, such as residing in urban areas, history of previous miscarriage, history of stillbirth, drinking unboiled water, and keeping a cat were found to have a non-statistical increased risk of being IgG seropositive, as shown in Table 2. Only low gestation age (OR 1.11; 95% CI, 1.02–1.20; *P*=.02) was found to predict IgG seropositivity on multivariate logistic regression analysis (Table 2).

Regarding the factors associated with IgM seropositivity, out of 169 women reported to keep cats, 5 (5.7%) were found to be IgM seropositive compared to only 1 out of 170 who reported not to keep cats (*P*=.037). Other factors such as history of stillbirth, drinking unboiled water, and not eating mutton or chicken were found to have a non-statistical increased risk of IgM seropositivity, as shown in Table 3. Only keeping cat (OR 11.8; 95%CI, 1.32–10.5; *P*=.03) was found to predict IgM seropositivity on multivariate logistic regression analysis (Table 3).

FIGURE 1. Graph of Prevalence of *T. gondii* IgG Antibodies by Gestational Age



Histopathological Changes of Conceptus Products and *T. gondii* IgG Seropositivity

Conceptus products were obtained from 171 women. The histopathological changes examined included presence/absence of inflammation, including inflammatory cells such as macrophages, neutrophils, lymphocytes, and eosinophils, calcification, necrosis, and presence of tachyzoites and bradyzoites (Figures 2 and 3).

Presence of inflammation (OR 1.95; 95% CI, 1.05–3.64; $P=.04$), calcification (OR 3.28; 95% CI, 1.01–10.63; $P=.05$), necrosis (OR 2.86; 95% CI, 1.39–5.89; $P=.01$), and lymphocytes (OR 2.24; 95% CI, 1.17–4.24; $P=.01$) were significantly associated with specific *T. gondii* IgG seropositivity (Table 4). The presence of histopathological changes had a non-statistical association with increased IgG median titres.

DISCUSSION

T. gondii infections have been implicated in poor pregnancy outcomes and as a common cause of cerebral toxoplasmosis among patients infected with HIV. For the first time in Tanzania, we document a high seropositivity of specific *T. gondii* IgG antibodies among women with spontaneous abortion. Our findings are comparable to those reported elsewhere.^{24,29} In comparison to previous studies in Sudan and Tehran,^{30,31} the reported prevalence in the current study is indeed high. The difference could be attributed to a number of factors, such as geographical variation and climatic conditions, which have been found to influence toxoplasmosis

worldwide.³² Moist and warm temperatures have been reported to enhance the sporulation of *T. gondii* oocysts. Generally, Mwanza's climate is warm with the temperature peaking around the third quarter of the year, which may explain high transmission rates. In addition, the prevalence reported in this study is significantly higher than what was reported in a previous study among pregnant women in the same setting and in an earlier report of data collected in the general population.^{20,22} The difference could be explained by the difference in study populations emphasizing the possible role *T. gondii* in causing spontaneous abortion.

The IgG seropositivity in this study was found to increase with an increase in maternal age, this confirms what was reported earlier.^{20,33} In addition, a lower gestation age was significantly associated with IgG seropositivity in this study. This corroborates the fact that most of the adverse *T. gondii* infection outcomes tend to occur during early pregnancy.¹² Furthermore, women with a previous history of miscarriage and a history of stillbirth had increased odds of being IgG seropositivity, which was also reported previously.^{31,34}

T. gondii IgM seropositivity in the current study is comparable to the previous study in Tehran, Iran, which reported a seropositivity of 2.7%.³¹ In contrast, the reported IgM seropositivity in the current study is low compared to previous studies in Sudan and Egypt.^{24,29,30} In comparison with a previous study conducted in the same settings²⁰ among pregnant women with full-term delivery, the IgM seropositivity in the current study is indeed high. The presence of specific *T. gondii* IgM antibodies among these women may explain the primary infection, which is often associated with poor foetal outcome when acquired in the first 12 weeks of the pregnancy. However, in many cases, specific *T. gondii* IgM antibodies tend to persist longer after primary infection. Therefore, in these cases, an IgG avidity test is recommended to exclude the possibility of previous infections.³⁶ Keeping cats was significantly associated with IgM seropositivity, which is consistent with other studies that reported the increased risk of *T. gondii* infections among pregnant women handling cats.^{33,37}

This study observed that the odds of being IgG seropositive were significantly high among women with placental histopathological changes. It has been previously suggested that the mechanism by which *T. gondii* induce abortion is through a chain of placental immunological reactions.²⁴ Animal model studies suggest that abortion can be induced by pathological changes, even without parasite replication in placental tissues.³⁸ Inflammation was significantly associated with IgG seropositivity in the current study. This finding has also been reported in a previous study with a positive correlation between intensity of inflammation and poor foetal outcome.³⁹ Despite the fact that placental inflammation is non-specific and that the majority of incidents are of unknown origin, a positive correlation with IgG seropositivity suggests that *T. gondii* infection might have played a key role in this context.^{40,41} Highly sophisticated techniques are recommended to confirm the presence of parasites in placental tissues, since

TABLE 2. Univariate and Multivariate Logistic Regression Analysis of Factors Associated With *T. gondii* IgG Seropositivity Among 260 Women With Spontaneous Abortion in Mwanza City

Characteristics (n)	IgG Seropositivity n (%)	Univariate OR (95% CI)	P-Value	Multivariate OR (95% CI)	P-Value
Age*	26±5.9	1.02 (0.98–1.07)	.188	1.03 (0.98–1.07)	.18
Gestation age*	11 (IQR 9–13)	1.09 (1.01–1.17)	.028	1.11 (1.02–1.2)	.02
Gravidity*	3 (IQR 1–4)	1.06 (0.92–1.22)	.397		
Residence					
Rural (160)	84 (52.5%)	1			
Urban (100)	60 (60%)	1.35 (0.81–2.25)	.237		
Education					
No formal/primary (147)	82 (55.8%)	1			
Secondary (86)	46 (53.5%)	1.00 (0.53–1.55)	.734		
Tertiary (27)	16 (59.2%)	1.15 (0.50–2.65)	.738		
Occupation					
Housewife (114)	57 (50%)	1			
Peasant (58)	37 (63.8%)	1.76 (0.92–3.37)	.087	1.54 (0.81–2.92)	.18
Employed (88)	50 (56.4%)	1.32 (0.75–2.30)	.336		
Marital status					
Single (38)	19 (50%)	1			
Married (222)	125 (56.3%)	1.23 (0.64–2.57)	.471		
Previous miscarriage					
No (221)	119 (53.9%)	1			
Yes (39)	25 (64.1%)	1.53 (0.75–3.09)	.237		
HIV status					
Negative (190)	105 (55.3%)	1			
Unknown (66)	36 (54.6%)	1.00 (0.55–1.77)	.920		
Positive (4)	3 (75%)	2.43 (0.24–23.77)	.446		
History of stillbirth					
No (225)	121 (53.8%)	1			
Yes (35)	23 (65.7%)	1.65 (0.78–3.47)	.189	1.51 (0.67–3.37)	.31
Keeping a cat					
No (170)	87 (51.2%)	1			
Yes (90)	57 (63.3%)	1.65 (0.97–2.78)	.062	1.29 (0.69–2.39)	.425
Unboiled water					
Yes (91)	56 (61.5%)	1			
No (169)	88 (52.1%)	1.47 (0.87–2.47)	.144		
Beef					
No (45)	25 (55.7%)	1			
Yes (215)	119 (55.4%)	0.99 (0.51–1.19)	.980		
Chicken					
No (84)	51 (60.7%)	1			
Yes (176)	93 (52.8%)	0.72 (0.42–1.23)	.233		
Mutton					
No (142)	85 (59.7%)	1			.235
Yes (118)	59 (50%)	0.67 (0.41–1.09)	.112	0.72 (0.43–1.23)	
Pork					
No (200)	105 (52.5%)	1			
Yes (60)	39 (65%)	1.68 (0.92–3.05)	.08	1.81 (0.95–3.44)	.06
Lamb					
No (215)	120 (55.8%)	1			
Yes (45)	24 (55.3%)	0.90 (0.47–1.72)	.761		

Abbreviations: CI, confidence interval; IgG, immunoglobulin G; IQR, interquartile range; OR, odds ratio.

TABLE 3. Univariate and Multivariate Logistic Regression Analysis of Factors Associated With *T. gondii* IgM Seropositivity Among 260 Women With Spontaneous Abortion in Mwanza City

Characteristics (n)	IgG Seropositivity n (%)	Univariate OR (95% CI)	P-Value	Multivariate OR (95% CI)	P-Value
Age*	22±3.7	0.85 (0.72–1.02)	.083	0.88 (0.68–1.13)	.319
Gestation age*	12 (IQR 11–12)	0.99 (0.77–1.27)	.946		
Gravidity*	1 (IQR 1–2)	0.56 (0.26–1.19)	.134	0.74 (0.28–1.96)	.548
Residence					
Rural (160)	4 (2.5%)	1			
Urban (100)	2 (2%)	0.79 (0.14–4.42)	.794		
Education					
No formal/primary (147)	3 (2.04%)	1			
Secondary (86)	2 (2.3%)	1.14 (0.18–6.97)	.885		
Tertiary (27)	1 (2.3%)	1.84 (0.18–18.43)	.602		
Occupation					
Housewife (114)	4 (3.5%)	1			
Peasant (58)	1 (1.74%)	0.48 (0.052–4.41)	.519		
Employed (88)	1 (1.14%)	0.31 (0.034–2.87)	.307		
Marital status					
Single (38)	1 (2.6%)	1			
Married (222)	5 (2.2%)	0.85 (0.96–7.5)	.886		
Previous miscarriage					
No (221)	5 (2.3%)	1			
Yes (39)	1 (2.5%)	1.13 (0.12–10)	.908		
History of stillbirth					
No (225)	5 (2.2%)	1			
Yes (35)	1 (2.9%)	1.29 (0.14–11.41)	.816		
Keeping a cat					
No (170)	1 (0.5%)	1			
Yes (90)	5 (5.7%)	9.94 (1.14–86.44)	.037	11.8 (1.32–105.04)	.027
Unboiled water					
Yes (91)	3 (3.3%)	1			
No (169)	3 (1.8%)	1.88 (0.37–9.54)	.443		
Chicken					
Yes (176)	2 (1.14%)	1			
No (84)	4 (4.8%)	4.54 (0.31–10)	.093		
Mutton					
Yes (118)	2 (1.7%)	1			
No (142)	4 (2.8%)	4.54 (0.30–10)	.553		
Pork					
Yes (60)	1 (1.7%)	1			
No (200)	5 (2.5%)	1.52 (0.17–14)	.708		
Lamb					
Yes (45)	1 (2.2%)	1			
No (215)	5 (2.3%)	1.05 (0.11–10)	.967		
Beef					
Yes (215)	2 (0.93%)				
No (45)	4 (8.9%)	10.4 (1.8–58.8)	.008		

* Beef and chicken have collinearity with cat so they were not fitted on multivariate logistic analysis. Abbreviations: CI, confidence interval; IgG, immunoglobulin G; IQR, interquartile range; OR, odds ratio.

FIGURE 2. (A) Hematoxylin and Eosin Stain (10x), Neutrophils Infiltrates Signifying Acute Inflammation. (B) Hematoxylin and Eosin Stain (20x), Lymphocytes and Plasma Cells Signifying Chronic Inflammation.

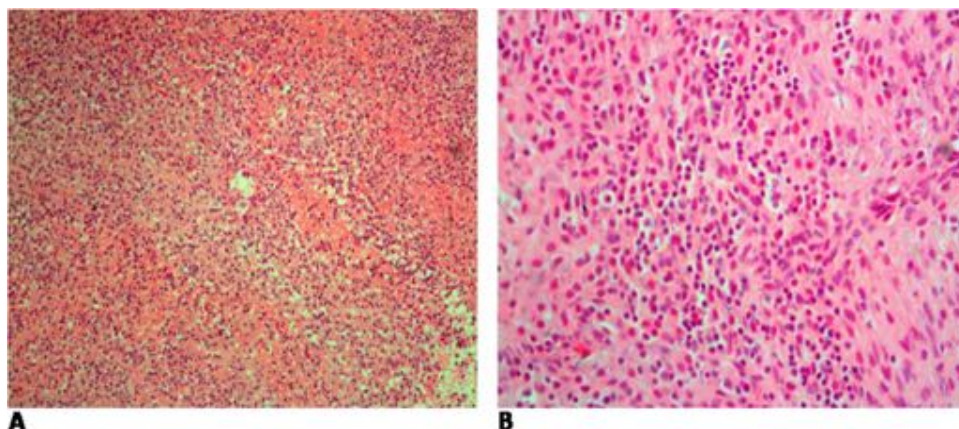
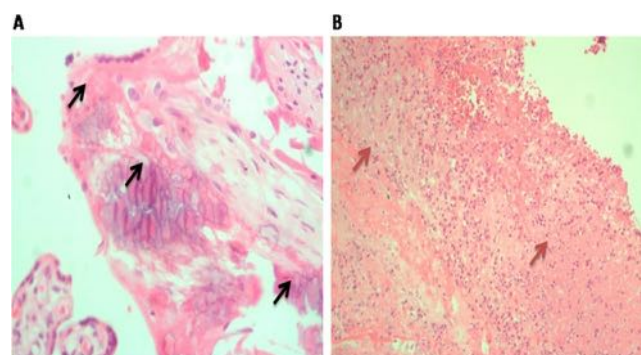


FIGURE 3. (A) Hematoxylin and Eosin Stain (20x), Black Arrows Showing Calcification. (B) Hematoxylin and Eosin Stain (10x) Brown Arrows Showing Areas of Necrosis with Neutrophils on the Background.



they can be easily overlooked in routine tissue studies. In addition, tissue necrosis and calcification have been found to be associated with IgG seropositivity in the current study, which is in agreement with a previous report.⁴² As a matter of fact, calcification and necrosis have been found to be common manifestations of toxoplasmosis.⁴³ Generally, a positive association between IgG seropositivity and pathological changes, as observed in this study, suggests that *T. gondii* infection might have played an important role in spontaneous abortion in our setting. Further studies on the mechanisms through which *T. gondii* infection causes abortion are needed to provide a clearer understanding.

TABLE 4. Histopathological Changes and IgG Seropositivity Among 171 Women With Spontaneous Abortion

Characteristics (n)	IgG Seropositivity	OR (95% CI)	P-Value
Inflammation			
Absent (103)	45 (43.7)	1	
Present (68)	41 (60.3)	1.95 (1.05–3.64)	.035
Calcification			
Absent (155)	74 (47.8)	1	
Present (16)	12 (75.0)	3.28 (1.01–10.63)	.047
Necrosis			
Absent (126)	55 (43.7)	1	
Present (45)	31 (68.9)	2.86 (1.39–5.89)	.004
Macrophages			
Absent (137)	64 (46.7)	1	
Present (34)	22 (64.7)	2.09 (0.95–4.56)	.064
Lymphocyte			
Absent (109)	47 (43.1)	1	
Present (62)	39 (63.0)	2.24 (1.17–4.24)	.014

Abbreviations: CI, confidence interval; IgG, immunoglobulin G; OR, odds ratio.

TABLE 5. Median Titres in Relation to Histopathological Changes Among 171 Women With Spontaneous Abortion

Characteristics (n)	Median IgG Titres (IU/mL)	IQR	P-Value
Inflammation			
Absent (45)	47.85	30.00–99.53	.252
Present (41)	75.93	30.00–202	
Calcification			
Absent (74)	54.22	27.19–154.38	.278
Present (12)	76.50	38.50–176.68	
Necrosis			
Absent (55)	52.22	30.00–118	.244
Present (31)	75.93	27.19–253	
Macrophages			
Absent (64)	54.22	30.54–118.18	.922
Present (22)	57.66	27.00–202.86	
Lymphocyte			
Absent (47)	52.22	30.00–99.77	.312
Present (39)	75.93	30.00–202.86	

Abbreviations: IgG, immunoglobulin G; IQR, interquartile range; IU, international units; mL millilitres.

Limitations

The major limitations of this study were the failure to perform IgG avidity. Furthermore, sensitive techniques like polymerase chain reaction and immunofluorescent techniques for antigen detection could provide more information on the existence of parasite DNA in placental tissues.

CONCLUSION AND RECOMMENDATIONS

The seropositivity of specific *T. gondii* IgG antibodies among women with spontaneous abortion and placental histopathological changes is alarmingly high in our setting, with a significant proportion of women at risk of contracting primary infection. The high prevalence of toxoplasmosis in women keeping cats, as one of the *T. gondii* IgM predictors, suggests that more education should be provided to these women on the risk of contracting *T. gondii* infection, especially during pregnancy. Ideally, these women should be excluded in activities that expose them to contact with infected cats or

cat faeces, such as feeding cats and gardening. This study has provided baseline information on the association between *T. gondii* infection and spontaneous abortion in a particular setting. As toxoplasmosis is one of the TORCH infections—toxoplasmosis, other (syphilis, varicella-zoster, parvovirus B19), rubella, cytomegalovirus, and herpes infections—implicated in causing poor pregnancy outcomes, these findings may contribute to the improvement of antenatal care services and may trigger policy makers into considering screening and treatment for the women found with a *T. gondii* infection during antenatal visits. In addition, our results emphasize the need to consider *T. gondii* infection as one of the possible causes of spontaneous abortion. Overall, a better understanding of the infection and its outcome, and the implementation of the measures mentioned above may have a dramatic impact on the reduction of the adverse pregnancy outcomes associated with *T. gondii* infection.

Acknowledgements: The authors would like to acknowledge the technical support provided by Mr. Vitus Silago and Mrs. Caroline Minja. Sincere gratitude to all staff at the Bugando Medical Centre, Sekou Toure regional hospital, Nyamagana district hospital, Buzuruga health centre, and Kamanga hospital. This study was supported by a research grant from CUHAS and BMC.

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Peer Reviewed

Competing Interests: None declared.

Received: 14 Dec 2016; **Accepted:** 25 Jul 2017

Cite this article as: Machumi I, Mirambo MM, Ruganuzza D, Rambau P, Massinde AN, Kihunrwa A, et al. Factors Associated With *Toxoplasma gondii* IgG and IgM Antibodies, and Placental Histopathological Changes Among Women With Spontaneous Abortion in Mwanza City, Tanzania. *East African Health Res J*. 2017;1(2):86–94. <https://doi.org/10.24248/EAHRJ-D-16-00408>

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TORCH Antibodies Among Pregnant Women and Their Newborns Receiving Care at Kilimanjaro Christian Medical Centre, Moshi, Tanzania

Aliasgher M Saajan,^{a,b} Mramba Nyindo,^{a,b} Joshua G Gidabayda,^{a,b} Mohammed S Abdallah,^{a,b} Shaneabbas H Jaffer,^b Aliasgher G Mukhtar,^b Tima M Khatibu,^b Rune Philemon,^{a,b} Grace D Kinabo,^{a,b} Blandina T Mmbaga^{a,b,c}

^aDepartment of Paediatrics and Child Health, Kilimanjaro Christian Medical Centre, Moshi, Tanzania; ^bKilimanjaro Christian Medical University College, Moshi, Tanzania; ^cKilimanjaro Clinical Research Institute, Moshi, Tanzania.
Correspondence to Aliasgher M Saajan (aliasghersajan@gmail.com).

ABSTRACT

Background: Toxoplasmosis, other (syphilis, varicella-zoster, parvovirus B19, and hepatitis B), rubella, cytomegalovirus (CMV), and herpes simplex virus type 1 and type 2 (HSV-1 and HSV-2) – known by the acronym TORCH – is a group of infections affecting both mothers and their unborn babies with adverse short- and long-term outcomes. The majority of infected mothers are asymptomatic, which leaves only speculation as to the probable cause of many congenital anomalies, stillbirths, prematurity, and death resulting from TORCH infections. The main objective of this study was to investigate previous exposure to TORCH infections by measuring the seroprevalence of TORCH antibodies in pregnant women and their newborns receiving care at Kilimanjaro Christian Medical Centre (KCMC), Moshi, Tanzania.

Methods: This was a cross-sectional, hospital-based study conducted at KCMC from December 2013 to April 2014. Of 350 pregnant women enrolled in the study, we tested 347 pregnant women attending the antenatal clinic and who opted to deliver at KCMC. Cord blood was collected and analysed for 309 of their newborns. To identify immunoglobulin G (IgG) and immunoglobulin M (IgM) antibodies in mothers and IgM antibodies in newborns, we used enzyme-linked immunosorbent assay testing. A structured questionnaire was used to collect data of mothers and their newborns. Data analysis was done using SPSS version 20.

Results: The seroprevalence of IgG antibodies to TORCH infections among pregnant women was 154 (44.4%) for toxoplasmosis, 311 (89.6%) for rubella, 343 (98.6%) for CMV, and 346 (99.7%) for HSV-1 and HSV-2; 141 (40.6%) had been exposed to all 4 infections. For HSV-1 and HSV-2, the IgM antibodies were found in 137 (39.5%) of the 347 pregnant women included in this study. Age above 35 years (OR 6.15; 95% CI, 1.22–31.1; $P=0.028$) and multiparity (OR 1.63; 95% CI, 1.01–2.62; $P=0.045$) were associated with higher risk of being exposed to all TORCH infections. A total of 11 newborns had IgM antibodies to HSV-1 and HSV-2 giving a seroprevalence of 3.6%, and one newborn had IgM antibodies to rubella, giving a seroprevalence of 0.3%. None of the newborns had antibodies to toxoplasmosis and CMV.

Conclusions: Exposure to TORCH infections was high among pregnant women in our population. Older age and multiparity were associated with a higher risk of being exposed to all TORCH infections. Seroprevalence to HSV-1 and HSV-2 was high in newborns. The higher IgM antibodies to HSV-1 and HSV-2 among pregnant mothers and their newborns may disturb maternal, fetal, and neonatal health, and therefore we recommend establishing treatment protocol to support management of pregnant women and newborns who are seropositive for IgM antibodies.

INTRODUCTION

A wide range of microorganisms including bacteria, viruses, fungi, and protozoans may infect a

pregnant woman and can lead to fetal death, organ injury, or short- and long-term sequelae depending on the offending pathogen.¹ The classical group of microorganisms is known as TORCH, which includes

toxoplasmosis, other (parvovirus B19, varicella-zoster virus infection, syphilis, hepatitis B), rubella, cytomegalovirus (CMV), and herpes simplex virus types 1 and 2 (HSV-1 and HSV-2).²

These organisms acquired in utero can lead to resorption of the embryo, abortion, stillbirth, malformation, intrauterine growth restriction, prematurity, or sequelae of chronic postnatal infection.³ Infection acquired during the intrapartum or early postpartum period may result in severe systemic disease that leads to death or persistent postnatal infection.

In most cases, maternal illness due to TORCH infections is mild, but the impact on the developing fetus is more severe.⁴ Clinical evidence of TORCH infections may be seen at birth, soon afterwards, or not until weeks, months, or years later. This is exemplified by a study of a large cohort of newborns with congenital toxoplasmosis in Brazil, which showed high rates of early retinochoroidal lesions (~80%) and active lesions (~50%).⁵ Prevalence of toxoplasmosis based on the detection of immunoglobulin G (IgG) antibodies was reported to be 30.9% in the Mwanza Region of Tanzania.⁶ Worldwide, overall risk of transmission for congenital toxoplasmosis is reported to be 30% and increases with gestational age at maternal seroconversion, from less than 15% at 13 weeks of gestation to almost 71% at 36 weeks of gestation. Prevalence of congenital infection ranges from 0.1 to 0.3 per 1,000 live births.⁷ The classic triad of signs suggestive of congenital toxoplasmosis includes chorioretinitis, hydrocephalus, and intracranial calcifications.⁸

Worldwide, CMV seroprevalence among women of reproductive age ranges from 45% in higher-income countries to 100% in lower- and middle-income countries.⁹ According to a recent review of 11 studies, CMV seroprevalence of adolescents is 90% and >95% during early adulthood and the average transmission rate is 0.65%, ranging from 0.6% in Panama to 6.1% in China.¹⁰ The same review showed a range of 0%–29% classified as symptomatic CMV at birth.¹⁰ Another review of 15 studies found that long-term sequelae from congenital CMV occurred 3 to 4 times more in symptomatic infants (40%–58%) compared with asymptomatic infants (13.5%). More children with long-term sequelae from congenital CMV were asymptomatic at birth.¹¹

In a study from 2011, CMV had been detected in about 20% of children in daycare centres, but CMV was also detected in about 10% of children who were not in daycare centres.¹² In a 2017 study in China, a seroprevalence of 96.2% among pregnant women was reported with a CMV transmission rate ranging from 0.4% to 0.7% depending on the specimen screened.¹³ In a study from Japan in 2006, congenital CMV was directly responsible for a substantial proportion of early childhood sensorineural hearing loss, and almost half of the infants at risk for the development of late onset CMV or gap junction beta-2 protein associated sensorineural hearing loss showed no clinical or audiological indications at birth.¹⁴

Screening data from the Herpevac Trial for Women revealed that half (51%) of participants screened had antibodies for HSV-1 (with or without HSV-2) and 11% had HSV-2 antibodies with or without HSV-1.¹⁵ The prevalence of antibodies to HSV-1 and HSV-2 increased with age, between the ages of 18 and 30. In Tanzania, HSV-2 seroprevalence was 80% among women at high risk in north-western Tanzania,¹⁶ 20.7% among pregnant women in the rural Manyara and Singida regions,¹⁷ and 35% in Dar es Salaam.¹⁸ Neonatal HSV infection is acquired during 3 distinct periods: intrauterine (in utero 5%), peripartum (during labour and delivery 85%), and postpartum (postnatal 10%).¹⁹ Intrauterine infection is associated with severe HSV infection regardless of the timing of the acquisition during gestation; at birth, it is characterized by a triad of findings, including skin vesicles or scarring, eye damage, and severe manifestations of microcephaly or hydranencephaly.²⁰ Data from the United States showed substantial utilization of resources for neonates with HSV. The median hospital charge was US\$37,431 (interquartile range US\$14,667–US\$74,559) per infant.²¹ The financial burden for congenital rubella syndrome is also substantial, estimated at US\$4,200 to US\$57,000 per case annually in middle-income countries and up to US\$140,000 over a lifetime in high-income countries.²²

Given the unfolding epidemiology, limited data, severe complications, and high economic burden of TORCH infections in the care of neonates with 1 or more of these infections, it is critical to thoroughly survey the distribution of TORCH infections in northern Tanzania. TORCH infections pose a substantial public health problem because the infected mothers are mostly asymptomatic, but the infections can lead to death, organ injury, or severe short- and long-term sequelae for their unborn fetuses and newborns. Thus, we aimed to study the immune status to congenital infections by TORCH agents among pregnant women and their newborns in northern Tanzania.

METHODOLOGY

Study Design

This was a cross-sectional study conducted from December 2013 to April 2014 at the Kilimanjaro Christian Medical Centre (KCMC), in Moshi, northern Tanzania. The study was conducted in the Obstetrics and Gynaecology Department and Paediatrics and Child Health Department using the antenatal clinic, labour ward, and neonatal ward. KCMC is a faith-based institution, primarily serving patients from the Kilimanjaro Region and neighbouring regions, such as Arusha, Manyara, Singida, and Tanga, as well as other parts of Tanzania. The number of hospital deliveries range from 3,500 to 4,000 per year, with more than half from the Moshi urban area and nearly 20% referred for medical reasons.²³

Study Population

All pregnant women above the age of 18 who attended the antenatal clinic during the study period, had a gestational age of at least 28 weeks (according to last menstrual period), and gave voluntary consent were enrolled in the study (n=350). Pregnant women not planning to deliver at KCMC and those with emergency referrals for delivery were excluded. Of the 350 pregnant women enrolled in the study, 312 delivered at KCMC, 10 delivered at nearby health centres, and 28 were lost to follow-up (Figure 1).

Sample Size Estimations

The minimum sample size was estimated using a formula by the Survey System software package (1988), expressed as $[Z^2 \cdot (p) \cdot (1-p)] / c^2$, where $Z=1.96$ for 95% confidence level (CI). A prevalence percentage (p) of 35% was selected based on a study done in Dar es Salaam¹⁸ and c represented the minimal tolerable error at 95% CI, expressed as a decimal (.05). The minimum estimated sample size was 344 participants. The study therefore recruited 350 pregnant women and their infants (mother-infant pairs).

Data Collection

We collected sociodemographic, behavioural, clinical, and delivery data of mothers enrolled in the study using pre-tested data collection tools. Data collected included age, level of education, marital status, alcohol consumption, parity, mode of delivery, and related maternal and fetal outcomes.

After enrolment, a laboratory request form was administered. Approximately 4 ml of whole blood was collected from each participant using BD Vacutainer blood collection red-top tubes (BD Medical, Plymouth, UK). Blood was allowed to clot for 1 hour and transported to Kilimanjaro Christian Research Institute – a biotechnology laboratory situated within the KCMC campus. Centrifugation was done at 1000–1300 g for 10 minutes. The supernatant (ie, serum) was transferred into cryotubes and stored in a refrigerator at -70°C for later analysis. All newborns were followed up immediately after delivery and 2 ml of cord blood were collected in BD Vacutainer blood collection red-top tubes for both liveborn and stillborn babies. The blood samples were processed and stored as described earlier.

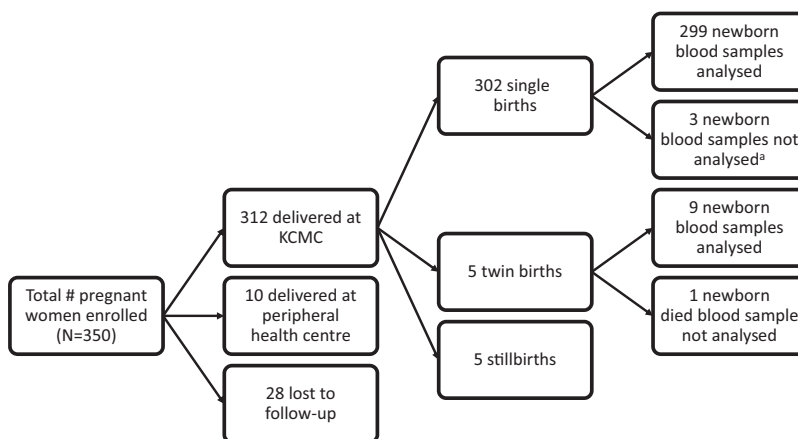
Laboratory Analysis

Laboratory analysis was done at the Kilimanjaro Christian Research Institute. All blood samples and kit reagents were brought to room temperature (23°C–25°C) before sample analysis was done. This was followed by testing for antibodies (IgG and IgM) to toxoplasmosis, rubella, CMV, HSV-1, and HSV-2 using an enzyme-linked immunosorbent assay (ELISA) test kit for mothers and only immunoglobulin M (IgM) antibodies to TORCH for newborns, according to the manufacturer’s instructions, as used for serum testing in this study.

Assay Procedure for TORCH IgG

Ten microlitres (10 µl) of blood sample were diluted with 1 ml of sample diluent. For each standard, 100 µl of diluted

FIGURE 1. Number of Participants Recruited and Newborn Blood Samples Analysed



Abbreviations: KCMC, Kilimanjaro Christian Medical Centre.

^aThe sample size was not sufficient and therefore was not analysed.

test sera were added into microplate wells and covered with cardboard sealer. The microplate wells were incubated for 30 minutes at room temperature (22°C–28°C). The plate covers were removed and contents discarded. The microplate wells were washed 5 times (each with 300 µL of working wash solution). A 100 µL of conjugate solution was added into all wells and covered with cardboard sealer. The microplate wells were incubated for 30 minutes at room temperature (22°C–28°C). The plate covers were removed and contents discarded. The microplate wells were washed 5 times. Each well was mixed with 100 µL of chromogenic substrate solution and then incubated at room temperature in a dark room for 15 minutes. To stop the reaction, 100 µL of stop solution was added to the wells. Absorbance was recorded at 450 nm by an ELISA reader.

Assay Procedure for TORCH IgM

We added 100 µL of each control as well as diluted test sera into appropriate wells. Two consecutive wells in the first strip were considered blank and positive, respectively, and the next 2 wells for duplicate negative control serum. The microplate wells were covered with cardboard sealer tightly and incubated for 30 minutes at 37°C. The plate covers were removed and contents discarded. The microplate wells were washed 5 times, each with 300 µL of working wash solution. A 100 µL conjugate solution was then added into all wells except the blank well. The microplate wells were covered with cardboard sealer tightly and incubated for 30 minutes at 37°C. The plate covers were removed and contents discarded. The microplate wells were washed 5 times. Each well was mixed with 100 µL of chromogenic substrate solution and then incubated at room temperature in a dark room for 15 minutes. To stop the reaction, 100 µL of stop solution was added to the wells. Absorbance was recorded at 450 nm by an ELISA reader.

Validation of serological assays was done and the cut-off values for detection of IgG and IgM were calculated according to the manufacturer's instructions.

Result Calculation and Evaluation for TORCH IgG

To distinguish between positive and negative results, the serum/cut-off ratio (S/Co) index was used and calculated according to manufacturer's index cut-off value for this ELISA:

$$S/Co = \frac{\text{sample optical density (OD)}}{\text{cut-off value}} \\ (\text{Cut-off value} = 10 \text{ antibody units per ml standard mean OD})$$

Based on this index, results higher than 1.1 were considered positive and results less than 0.9 were considered negative.

Result Calculation and Evaluation for TORCH IgM

To distinguish between positive and negative results, the cut-off index was determined by using the following formula as per the manufacturer manual:

$$\text{Cut-off index} = \frac{\text{OD of sample}}{\text{cut-off value}} \\ (\text{Cut-off value} = \text{mean OD of negative control serum} + 0.15)$$

Statistical Analysis

Data were analysed using IBM SPSS Statistics v. 20 (Armonk, New York, USA) where descriptive statistics were estimated. For combined TORCH infection, univariate analysis was done to estimate the odds ratios and the 95% confidence interval to determine factors associated with exposure to both TORCH infections with $P < .05$ used as a cut-off value to indicate statistical significance.

Ethical Consideration

Ethical clearance was obtained from the Kilimanjaro Christian Medical University College via the College Research Ethics Review Committee and permission was obtained from KCMC Obstetrics and Gynaecology Department and Paediatrics and Child Health Department. Assent to participate and informed written consent was sought from each pregnant woman prior to involvement in the study.

RESULTS

Sociodemographic Characteristics of the Study Participants

The mean age of the pregnant women was 29 (standard deviation [SD]=5.1) years at enrolment, 191 participants (54.6%) were between 21 and 30 years old, and 201 (57.4%) participants were married. Most of the participants (74.9%) had attended secondary education and the majority of them were living in urban areas (89.1%). Table 1 shows the socio-demographic characteristics of the study participants.

Behavioural, Clinical, and Delivery Characteristics of the Participants

About 15% of the study participants reported to have consumed alcohol during the current pregnancy (Table 2). A total of 12 (3.4%) pregnant women enrolled in this study were HIV positive. Fifty-nine percent tested negative for the venereal disease research laboratory syphilis test and 40.3% did not know their syphilis infection status. Regarding parity, 240 (68.6%) participants were multiparous. The mean gestational age was 38.2 weeks (SD=3.3). Regarding the mode of delivery, 187 (60.1%) participants underwent spontaneous vaginal delivery and single live birth was the most common delivery outcome (96.8%). No major post-delivery complications among mothers were reported. History of prolonged rupture of membrane was rare.

TABLE 1. Sociodemographic Characteristics of the Participants (N=350)

Characteristics	n	%
Age in years		
≤ 20	15	4.2
21–30	191	54.6
31–40	142	40.6
41+	2	0.6
Mean age in years (SD)	29.1 (5.1)	
Marital status		
Single	2	0.6
Married	201	57.4
Cohabiting	147	42.0
Level of education		
Primary education	88	25.1
Secondary education and above	262	74.9
Occupational status		
Unemployed	3	0.9
Self-employed	144	41.5
Employed	144	41.5
Housewife	36	10.4
Student	20	5.8
Residence		
Rural	38	10.9
Urban	312	89.1

Abbreviation: SD, standard deviation.

Seroprevalence of IgG and IgM Antibodies to TORCH in Pregnant Women

Overall, 141 (40.6%) pregnant women participants were seropositive for all 4 TORCH infections in our study. Ages above 35 years (OR 6.15; 95% CI, 1.22–31.19; $P=0.028$) and multiparity (OR 1.63; 95% CI, 1.01–2.62; $P=0.045$) were associated with a higher risk of seropositivity for all TORCH infections (Table 3).

Seroprevalence of IgG antibodies was as follows: toxoplasmosis 154 (44.4%), rubella 311 (89.6%), CMV 342 (98.6%), and HSV-1 and HSV-2 (99.7%) (Figure 2). Seroprevalence of IgM antibodies to HSV-1 and HSV-2 was 137 (39.5%) (Figure 3).

TABLE 2. Behavioural, Clinical, and Delivery Characteristics of Participants

Characteristics (n)	n	%
Drug abuse or addiction (350)		
Alcohol	54	15.4
None	296	84.6
HIV status (350)		
Positive	12	3.4
Negative	331	94.6
Unknown	7	2.0
Syphilis status (350)		
Positive	2	0.6
Negative	207	59.1
Unknown	141	40.3
Parity (350)		
Nulliparous	110	31.4
Multiparous	240	68.6
Mean age in years at gestation (SD)	34.2 (3.3)	
Place of delivery (350)		
Kilimanjaro Christian Medical Centre	312	89.0
Peripheral health centre	10	2.9
Lost on follow-up	28	8.0
Mode of delivery (311)		
Spontaneous vaginal delivery	187	60.1
Caesarean section	124	39.9
Pregnancy outcomes (311)		
Singleton live birth	301	96.8
Multiple live births	5	1.6
Stillbirth, macerated	2	0.6
Stillbirth, fresh	3	1.0

Abbreviation: SD, standard deviation.

Seroprevalence (IgM) Antibodies to TORCH in Newborns

Seroprevalence of IgM antibodies to HSV-1 and HSV-2 among newborns was 11 (3.6%). One newborn was co-infected with rubella, for a prevalence of 0.3% for rubella.

TABLE 3. Maternal and Clinical Characteristics of Participants with IgG Antibodies to TORCH Infections Present (N=347)

Characteristics	Total	All TORCH IgG		P-value
		n (%)	OR (95% CI)	
Age (years)				
≤ 20	15	2 (13.2)	Ref.	
21–35	295	121 (41.0)	4.52 (1.00–20.39)	.050
36+	37	18 (48.6)	6.15 (1.22–31.19)	.028
Marital status				
Single	2	1 (50.0)	1.81 (0.11–29.50)	.678
Married	199	88 (44.2)	1.43 (0.92–2.22)	.108
Cohabiting	146	52 (35.6)	Ref.	
Level of education				
Primary	86	36 (41.9)	1.07 (0.65–1.75)	.789
Secondary and above	261	105 (40.0)	Ref.	
Occupational status				
Unemployed	3	1 (33.3)	1.30 (0.11–16.0)	.838
Self-employed	144	67 (46.5)	2.26 (1.02–5.03)	.045
Employed	144	57 (39.6)	1.70 (0.76–3.80)	.193
Housewife	36	10 (27.8)	Ref.	
Student	20	6 (30.0)	1.11(0.34–3.71)	.860
Residence				
Rural	36	14 (38.9)	Ref.	
Urban	311	127 (40.8)	1.09 (0.54–2.20)	.822
Parity				
Nulliparous	107	35 (32.7)	Ref.	
Multiparous	240	106 (44.2)	1.63 (1.01–2.62)	.045
History of PROM				
Yes	10	3 (30.0)	Ref.	
No	298	122 (40.9)	1.62 (0.41–6.38)	.365
Mode of delivery				
SVD	188	71 (37.8)	Ref.	
C/S	122	56 (45.9)	1.40 (0.88–2.22)	.155
HIV status				
Positive	12	7 (58.3)	2.05 (0.64–6.60)	.228
Negative	328	133 (40.5)	Ref.	
Unknown	7	1 (14.3)	0.24 (1.03–2.05)	.194
Syphilis status				
Positive	2	2 (100.0)	2.86	.999
Negative	205	74 (36.1)	Ref.	
Unknown	140	65 (46.1)	1.53 (0.99–2.38)	.055

Abbreviations: C/S, caesarean section; PROM, premature rupture of membrane; Ref., reference group; SVD, spontaneous vaginal delivery.

FIGURE 2. Prevalence of TORCH IgG Antibodies Among Participants, by Co-Infection

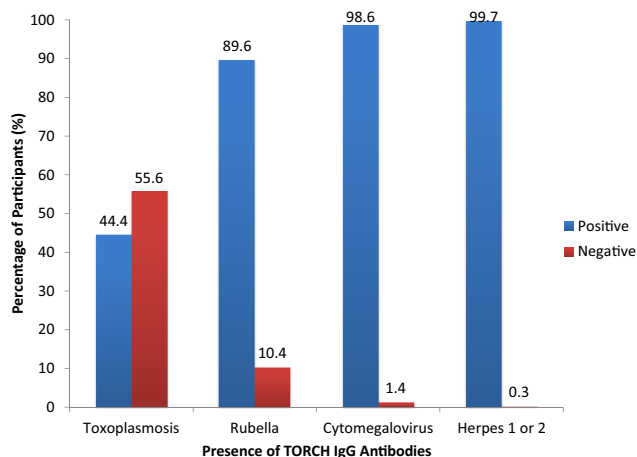


FIGURE 4. Prevalence of TORCH IgM Antibodies Among Newborns, by Co-Infection

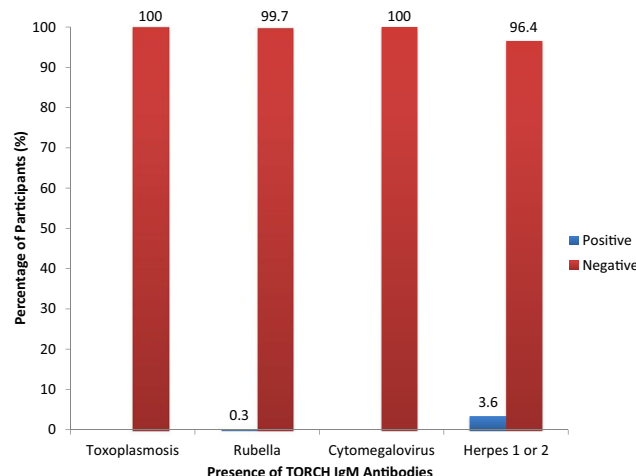
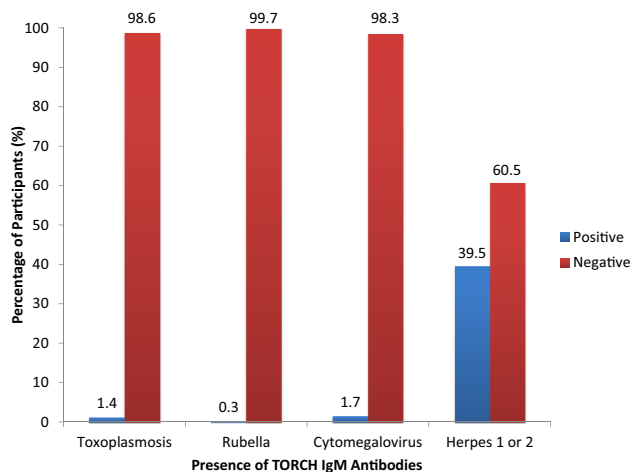


FIGURE 3. Prevalence of TORCH IgM Antibodies Among Participants, by Co-infection



The mothers of 9 of the 11 newborns had previously been exposed to all TORCH infections investigated in this study: toxoplasmosis, rubella, CMV, and HSV-1 and HSV-2. All newborns who tested positive for IgM antibodies were single births, born with Apgar scores above 7, had no reported birth trauma or birth defects, had HIV-negative mothers, lived in urban areas, and their mothers had no history of premature

rupture of membranes during labour. None of the newborns were infected with toxoplasmosis or CMV (Figure 4).

DISCUSSION

The objective of this study was to investigate previous exposure to TORCH infections among pregnant women and their newborns receiving care at KCMC in Moshi, Tanzania, by measuring their levels of antibodies to the infections. Our findings showed a high seroprevalence of IgG antibodies to toxoplasmosis, rubella, CMV, and HSV-1 and HSV-2. A high seroprevalence of IgM antibodies to HSV may indicate ongoing infection, which increases risk of HSV transmission to the newborn. We also found that 3.6% of the newborns born to mothers with HSV tested positive for IgM antibodies, which indicates maternal intrauterine transmission. Overall, 40.6% of pregnant women were seropositive for all TORCH infections in this study. Higher exposure to these infections may therefore lead to concurrent infection, which could increase the chances of transmission, as observed in 9 of 11 newborns in this study who were HSV-positive and born to mothers who tested positive for IgG antibodies to all TORCH infections in this study. The higher IgG seroprevalence observed in this study in – which nearly all women were previously exposed to some TORCH infections – may indicate that women of reproductive age are exposed to the infections before pregnancy in the general population. This may support generalizability of the study findings; however, because it was a highly selective population of study participants in a tertiary care hospital, we must be cautious about generalizing the findings.

Previous exposure to toxoplasmosis among pregnant women in our study was high, which could be attributed to geographical conditions, personal hygiene, eating habits, and lack of awareness of the disease before conception. Similar findings of seroprevalence for toxoplasmosis were reported in Colombia (45.8%)²⁴ and Albania (48.6%).²⁵ On the other hand, our findings showed a higher seroprevalence for TORCH infections than in Mexico (8.2%),²⁶ the United Kingdom (9.1%),²⁷ Japan (10.3%),²⁸ Burkina Faso (20.3%),²⁹ Palestine (27.9%),³⁰ Croatia (29.1%),³¹ Kosovo (29.4%),³² Nigeria (32.6%),³³ and Iran (37.2%).³⁴ More-over, our findings showed a higher seroprevalence than recently described by Mwambe and colleagues in Tanzania (30.9%).⁶ Comparatively, our findings show a lower prevalence than reported in Saudi Arabia (51.4%)³⁵ and some parts of Brazil (59.8% in Palotina and 60.6% among Jesuits).³⁶ The variation of prevalence in different countries and regions may be attributed to geographical conditions, personal hygiene, eating habits, and lack of awareness of the disease. In our study, no newborn tested seropositive for antibodies to toxoplasmosis. The majority of newborns infected during pregnancy are asymptomatic at birth; however, 80% of them may develop long-term learning and visual disabilities later in their lives.³⁷

Exposure to rubella before conception among pregnant women in this study was high, which may indicate a heavy circulation of wild-type rubella viruses in our population because widespread vaccination is not yet available in Tanzania.³⁸ Moreover, infection of rubella virus leads to lifelong immunity. Similar trends of exposure to rubella were reported in Nigeria (87.5%),³⁹ Tanzania (92.9%),⁴⁰ Haiti (93.4%),⁴¹ Turkey (93.8%),⁴² and Croatia (94.6%)³¹; however, our findings showed a higher prevalence to rubella than reported in Burkina Faso (77%)⁴³ and western Sudan (65.3%).⁴⁴

Exposure to CMV among pregnant women was also high in our study. The risk of CMV exposure to women of reproductive age increases when their child attends daycare¹² and the fact that the majority of participants in our study were educated, employed, multiparous, and lived in urban areas, where the possibility of children attending daycare is high, may contribute to the high levels of exposure to CMV. Similarly high exposure rates were reported in Iran (97.7%),⁴⁵ Nigeria (97.2%),⁴⁶ Palestine (96.6%),⁴⁷ and Taiwan (91.1%)⁴⁸; however, lower rates of exposure were reported in Croatia (75.4%),³¹ Norway (62.8%),⁴⁹ and Tanzania (63.1%) from a study in 1990.⁵⁰

Additionally, exposure to HSV-1 and HSV-2 among pregnant women in the study was high. This could be explained by the changing epidemiology of HSV disease toward HSV type-1 more than type-2, changing sexual behaviour, and the latency of HSV post-primary infection. Similar findings have been reported in the Republic of Vanuatu (100%)⁵¹ and Croatia (85.5%)³²; however, lower exposures were reported in Norway (14%),¹⁸ Belgium

(18.2%),⁵² and in the Manyara and Singida regions of Tanzania (20.7%).¹⁷ Parallel findings were seen among women with high risk reported in northern Tanzania.¹⁶ The risk of transmission of HSV-1 and HSV-2 to newborns was only from mothers with antibodies (ie, IgG and IgM antibodies to HSV-1 and HSV-2). This may be explained by acute maternal infection or reactivation of latent infection.

HSV was the only one with higher transmission rate (3.6%), however, newborns were asymptomatic at birth and during the early neonatal period. A review of congenital herpes cases in the past 4 decades revealed that among 64 cases with clinical presentations, cutaneous lesions were the most common clinical manifestation where less than one-third of the cutaneous presentation had typical triad findings at birth, 44% had manifestations other than vesicles or bullae, 67% of patients had central nervous system manifestation and 39% patients had ocular findings, 18% of whom had retinal disease.⁵³ Neonatal HSV infection is treated with acyclovir, 60 mg/kg of body weight per day in 3 doses (20 mg/kg per dose) given intravenously. Disseminated and central nervous system infections are treated for at least 21 days.²⁰ Currently, no treatment protocol exists at KCMC for newborns with HSV infection; therefore, no treatment is given for newborns who test HSV positive.

Strengths and Limitations

This study had a large sample size that included both pregnant women and their newborns. We were therefore able to estimate antibodies for 4 infections belonging to the TORCH group of infections and the transmission rate, primarily for HSV-1 and HSV-2.

Despite its strengths, this study also had some limitations. Limitations included a highly selective population of study participants because it was a hospital-based study in a tertiary care hospital, and therefore our findings might not reflect exposure in the general population. Second, the loss to follow-up of nearly 10% of newborns in our study means that we do not know their serostatus.

CONCLUSIONS AND RECOMMENDATIONS

Exposure to TORCH infections was very high among pregnant women in our study population. Newborn seroprevalence of IgM antibodies to HSV-1 and HSV-2 was also high. HSV-1 and HSV-2 infections in pregnant women may disturb maternal, foetal, and neonatal health, and therefore, antenatal screening may be recommended. A large cohort study could help provide the evidence needed, including long-term sequelae for newborns who test positive for TORCH antibodies, in order to advocate for TORCH management during pregnancy and newborn care at KCMC.

HSV prevention advocacy to both partners during antenatal care is also recommended. Higher IgM antibodies to HSV-1 and HSV-2 among pregnant women and their

newborns indicate a need to establish treatment protocol to support management of pregnant women and newborns who are seropositive for IgM antibodies.

Acknowledgements: We appreciate the Medical Education Partnership Initiative (MEPI) from the Kilimanjaro Christian Medical University College and the Duke Global Health Institute for funding support through the HRSA award T84HA21123. Our appreciation to the former Head of Department of Obstetrics (Dr. Gilleard Massenga) who is currently the hospital Executive Director, all nurses and staffs at the Reproductive and Child Health Clinic, Obstetrics and Gynaecology Department, and Paediatrics and Child Health Department for support given during data collection. Finally, we would like to acknowledge the mother-infant pairs who agreed to participate in this study, and for their valuable information and cooperation throughout the study.

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Peer Reviewed
Competing Interests: None declared.

Received: 27 Oct 2016; **Accepted:** 1 Aug 2017

Cite this article as: Saajan AM, Nyindo M, Gidabayda JG, Abdallah MS, Jaffer SH, Mukhtar AG, et al. TORCH Antibodies Among Pregnant Women and Their Newborns Receiving Care at Kilimanjaro Christian Medical Centre, Moshi, Tanzania. *East African Health Res J*. 2017;1(2):95-104. <https://doi.org/10.24248/EHRJ-D-16-00340>

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Knowledge, Attitudes, and Perceived Risks Related to Diabetes Mellitus Among University Students in Uganda: A Cross-Sectional Study

Brenda Kharono,^a Ruth Nabisere,^a Nabyonga Kiddu Persis,^b Jackie Nakakeeto,^c Abraham Openy,^d Sabrina Bakeera Kitaka^{a,e}

^aSchool of Medicine, College of Health Sciences, Makerere University, Kampala, Uganda; ^bMbarara University of Science and Technology, Kampala, Uganda; ^cSchool of Health Sciences, Kampala International University, Ishaka, Uganda; ^dGulu University, Gulu, Uganda; ^eDepartment of Paediatrics and Child Health, Mulago Hospital, Kampala, Uganda.

Correspondence to Brenda Kharono (kharonob88@yahoo.co.uk).

ABSTRACT

Background: Diabetes mellitus is on the rise in low-income countries, including Uganda, owing to the ‘westernization’ of individual lifestyles. It remains unanswered whether the majority of university students who are rapidly embracing ‘western’ lifestyles have any knowledge of diabetes or perceive themselves to be at risk of acquiring the disease. The aim of the study was to assess the knowledge, attitudes, and perceived risks related to diabetes mellitus among university students in Uganda.

Methods: This descriptive cross-sectional study was conducted in 4 universities in Uganda from August to November 2013. The data collection tool included questions on risk factors, symptoms, personal risks, and practices to prevent diabetes mellitus. We interviewed 378 university students using pretested self-administered semi-structured questionnaires. Only students who consented to participate in the study were included. Data were entered into EpiData version 3.1 and analysed using SPSS version 18.

Results: Almost all (99%) of the students had knowledge about diabetes mellitus. The majority (83.1%) reported that diabetes mellitus is not completely a genetic/hereditary disease. Only a minority of respondents reported that they should worry about diabetes before 45 years of age. Common symptoms of diabetes reported by the respondents included constant hunger, blurred vision, fatigue, and frequent urination.

Conclusions: Our study revealed that the majority of university students in Uganda had good knowledge about the risk factors and symptoms of diabetes mellitus. The majority also perceived themselves to be at risk of diabetes.

BACKGROUND

Diabetes mellitus is a group of metabolic disorders sharing the common underlying feature of hyperglycemia.¹ The estimated worldwide prevalence of diabetes mellitus among adults was 382 million (8.3%) in 2013,² and is predicted to rise to around 438 million (7.7%) by 2030.³ There is likely to be an increase in the number of people living with diabetes mellitus worldwide unless preventive action is taken. Sub-Saharan Africa is reported to be one of the regions with the fastest growing rates of diabetes mellitus in the world. Global estimates anticipate the number of people affected by diabetes to increase by 98% from 12 million (3.8%) in 2010 to 24 million (4.7%) in 2030^{2,3}; and in Uganda,

the prevalence is expected to rise from 2.2% in 2010 to 3.1% by 2030.³

The American Diabetes Association defines diabetes as a group of metabolic diseases characterised by hyperglycemia, which results from defects in insulin secretion, insulin action, or both. It further classifies diabetes mellitus as either type 1 and type 2.⁴ Type 2 diabetes mellitus is the form of diabetes that results from a combination of resistance to insulin action and an inadequate compensatory insulin secretory response.⁴ Although type 2 diabetes is generally considered a disease occurring primarily in adults, it is now being diagnosed more frequently among the youth.⁵ In developing countries, the people in the middle, productive age of their lives are

particularly affected by diabetes. In these countries, about 75% of all people with diabetes are under 65 years old and 25% of all adults with diabetes are younger than 44 years. In contrast, more than half of all people with diabetes in developed countries are older than 65 and only 8% of adults with diabetes are younger than 44 years.⁵

The increase in diabetes mellitus in developing countries like Uganda is triggered by many factors, including the unhealthy diets that encompass consumption of high calories, smoking, alcohol use, and sedentary lifestyles, all of which have been subsequent to increasing urbanisation and socioeconomic development.^{6,7} Similar to other African countries, little effort has been invested in the prevention and management of non-communicable diseases (NCDs) in Uganda, despite an increase in the burden of NCDs in the sub-Saharan region.⁸ NCDs are chronic diseases (tend to be of long duration or for life) and are the result of a combination of genetic, physiological, environmental, and behavioural factors.⁸ The burden of infectious diseases like malaria and HIV/AIDS has overshadowed the 'silent' increase of NCDs, diabetes inclusive.^{6,8,9} The World Health Organization reported that over a period of 30 years, the burden of NCDs for developing countries was expected to rise by over 60% by 2020, compared with fewer than 10% in developed countries.^{10,11} The 'silent increase' in morbidity due to NCDs is attributed to a lack of knowledge and risk perception of the diseases in question.⁸

The cost of treatment for diabetes mellitus is high; the drugs are fairly expensive and the nature of disease demands long-term treatment. The focus is increasing on the prevention, detection, and effective treatment of diabetes. A high awareness of disease in a population is always instrumental in influencing the behaviour of people, as they could easily perceive themselves to be at risk, and thus work towards avoiding catching the disease in question.⁹ In Uganda, diabetes mellitus is mainly managed by doctors. Medical students participate in community-based education services, and are, therefore, expected to possess basic diabetes knowledge to pass on to their patients and the community.

There is no available literature about the knowledge, attitudes, and perceived risk related to diabetes mellitus among university medical students in Uganda. Because little information is known about the extent to which they perceive themselves to be at risk or how knowledgeable they are about the disease, this study aims to assess the knowledge, attitudes, and risk perceptions of university medical students in Uganda regarding diabetes mellitus.

METHODOLOGY

This descriptive cross-sectional study was conducted from 18 to 30 November 2013 among students in 4 Ugandan universities – Makerere University, Mbarara University of

Science and Technology, Kampala International University, and Gulu University Faculty of Medicine.

Inclusion and Exclusion Criteria

Participants were eligible if they had a valid university student identity card and were medical students, 18 years of age and above, and willing to provide consent to participate in the study during the allocated data collection period. University staff, students with invalid university identity cards, and those unwilling to provide consent for participation were not eligible for inclusion in the study.

Questionnaire Development and Pilot Test

Following literature review, a student research team from the 4 universities pre-tested self-administered semi-structured questionnaires at Makerere University 2 weeks prior to the official start date of data collection. The pre-test was done to assess question variation, difficulty, and meaning, while at the same time testing respondent interest and attention. Co-investigators, assisted by 10 data collectors had 3-day training on the data collection procedure. All self-administered questionnaires were prepared in English. After Institutional Review Board approval from the different university institutions, a pilot study was done.

Study Population and Sampling Technique

Although a total of 385 participants was anticipated, only 378 participants consented and were available to participate in the study at the allocated time. The 378 students were selected by simple randomization. Information about the study was read to them by the data collectors before their written informed consent was sought. The self-administered questionnaires were given to the eligible participants in their respective halls of residence. The distribution was done at times that were convenient for the students and did not conflict with academic activities.

The sample size (385) was determined using the Leslie Kish formula. The sample size was calculated as $n = \frac{(z^2)pq}{d^2}$, where n was the sample size; z was the value that corresponds to the 95% confidence, which is 1.96 test statistic; p was the proportion of study participants in the population (0.5); $q(1-p)$ was the probability that the outcome did not occur; and d was the acceptable error to be committed (5%), therefore, $n = 385$. However, only 378 questionnaires were received from the respondents — giving us a response rate of 98.2% — so this number was used in analysis. The independent variables for analysis were year of study, age, and course, and the dependent variables were knowledge, attitudes, and risk perceptions related to diabetes mellitus.

The first part of the questionnaire addressed sociodemographic data, which included age, sex, course, and year of study. The second part addressed overall knowledge of diabetes mellitus, risk factors, and symptoms. The third part covered risk assessment through family history of diabetes

mellitus and predisposing lifestyles, such as eating fast foods, smoking, physical inactivity, and alcohol consumption. The fourth part assessed attitudes and risk perceptions towards acquiring diabetes mellitus.

Data Analysis

The 378 questionnaires were checked for completeness to ensure collection of quality data. Double data entry into EpiData version 3.1 (Odense, Denmark) was done. Data was then exported to SPSS version 18 (Armonk, New York, USA) for analysis. Data was analysed using frequency and percentages, and SPSS was also used to determine the variation of the independent variables.

Ethics Statement

The study was approved by the research and ethics committee of Makerere University. Only students who provided written consent for participation were included in the study. The informed consent form was approved by the research and ethics committee of Makerere University, which was signed by each student prior to participation.

RESULTS

Sociodemographic Characteristics of Respondents

Among the 378 study participants, 229 (60.6%) were male and 149 (39.4%) were female (Table 1). Almost two-fifths (39.2%) of the study participants were from Makerere University, and the remaining three-fifths were divided almost equally between Mbarara University (20.6%), Gulu University (20.6%), and Kampala International University (19.6%).

Students from all the years in the universities had an almost equal participation in the study. The age range of participants was 18–40 years and the mean age was 22 years. About two-thirds (n=248, 65.6%) of the participants reported not having a family member that had been diagnosed with diabetes mellitus, while one-third (n=129, 34.1%) reported positive history of diabetes mellitus.

Knowledge of Study Participants Regarding Diabetes Mellitus

From Table 2, almost all (99.2%) the students in all the universities across all years of study had heard about diabetes mellitus. Only 3 students (0.8%) reported not having heard about diabetes – 2 from Makerere University (Year 4) and 1 from Gulu University.

Of the participants, the majority (83.1%) reported that genetic factors were responsible for diabetes mellitus causation, about a tenth (11.9%) reported that diabetes mellitus is not a genetic disease, and a minority (5%) said they did not know if diabetes mellitus was genetic in origin.

TABLE 1. Sociodemographic Characteristics of Respondents (N=378)

Respondent Characteristics	Responses	
	n	%
University		
Gulu	78	39.2
KIU	74	19.6
MAK	148	20.6
MUST	78	20.6
Year of Study		
1	75	19.8
2	74	19.6
3	77	20.4
4	74	19.6
5	78	20.6
Gender		
Male	229	60.6
Female	149	39.4

Abbreviations: KIU, Kampala International University; MAK, Makerere University; MUST, Mbarara University of Science and Technology.

More than half of the students who participated in the study said that consuming fast foods from restaurants (73.8%), smoking (60.3%), physical inactivity (77.8%), and alcohol consumption (83.3%) can predispose you to getting diabetes.

The majority of students (n=325, 86%) demonstrated good knowledge about the common signs and symptoms of diabetes mellitus, this was demonstrated by participants correctly identifying at least 3 of the signs and symptoms of diabetes. Participants knew that diabetes mellitus may present as excessive hunger (82.5%), blurred vision (74.6%), and frequent urination (86.8%). Only 7 participants demonstrated absolute ignorance about the listed signs and symptoms of diabetes mellitus. Of these, 1 student (Year 2) responded ‘no’ to all the listed symptoms, 4 (3 from Year 1 and 1 from Year 3) did not fully answer that question, 1 (Year 1) responded ‘no’ to frequent urination and did not respond to the other symptoms, while 1 (Year 1) responded ‘no’ to constant hunger, blurred vision, and increased blurred vision. It was noted that this question had the biggest percentage of unanswered sections.

TABLE 2. Frequency and Percentage of Knowledge of the Study Participants Related to Diabetes Mellitus (N=378)

Variables	Yes n (%)	No n (%)	I do not know n (%)	Not answered n (%)
Have heard about DM	375 (99.2)	3 (0.8)	n/a	n/a
DM is preventable	323 (85.4)	44 (11.6)	11	n/a
DM is genetic	314 (83.1)	45 (11.9)	19 (5.0)	n/a
DM affects only older people	12 (3.2)	357 (94)	9 (2.4)	n/a
DM can be transmitted from one person to another	14 (3.7)	346 (91.5)	15 (4)	3 (0.8)
Knowledge on Signs and Symptoms				
Muscle cramps				
Year 1	25 (6.6)	31 (8.2)	n/a	19 (5.0)
Year 2	34 (9.0)	32 (8.5)	n/a	8 (2.1)
Year 3	26 (6.9)	31 (8.2)	n/a	20 (5.3)
Year 4	29 (7.7)	35 (9.3)	n/a	10 (2.6)
Year 5	25 (6.6)	42 (11.1)	n/a	11 (2.9)
Blurred vision				
Year 1	32 (8.5)	30 (7.9)	n/a	13 (3.4)
Year 2	51 (13.5)	17 (4.5)	n/a	6 (1.6)
Year 3	60 (15.9)	6 (1.6)	n/a	11 (2.9)
Year 4	67 (17.7)	6 (1.6)	n/a	1 (0.3)
Year 5	72 (19.0)	4 (1.1)	n/a	2 (0.5)
Constant hunger				
Year 1	48 (12.7)	9 (2.4)	n/a	18 (4.8)
Year 2	59 (15.6)	11 (2.9)	n/a	4 (1.1)
Year 3	63 (16.7)	7 (1.9)	n/a	7 (1.9)
Year 4	66 (17.4)	5 (1.3)	n/a	3 (0.8)
Year 5	76 (20.1)	1 (0.3)	n/a	1 (0.3)
Increased feeling of tiredness				
Year 1	50 (13.2)	11 (2.9)	n/a	14 (3.7)
Year 2	51 (13.5)	12 (3.2)	n/a	11 (2.9)
Year 3	65 (17.2)	4 (1.1)	n/a	8 (2.1)
Year 4	57 (15.1)	9 (2.4)	n/a	8 (2.1)
Year 5	59 (15.6)	15 (4.0)	n/a	4 (1.1)

Continued

TABLE 2. Continued

Variables	Yes n (%)	No n (%)	I do not know n (%)	Not answered n (%)
Frequent urination				
Year 1	53 (14.0)	13 (3.4)	n/a	9 (2.4)
Year 2	64 (16.9)	5 (1.3)	n/a	5 (1.3)
Year 3	65 (17.2)	7 (1.9)	n/a	5 (1.3)
Year 4	71 (18.8)	3 (0.8)	n/a	0 (0)
Year 5	75 (19.8)	3 (0.8)	n/a	0 (0)
Excessive sweating				
Year 1	29 (7.7)	24 (6.3)	n/a	22 (5.8)
Year 2	44 (11.6)	21 (5.6)	n/a	9 (2.4)
Year 3	41 (10.8)	19 (5.0)	n/a	17 (4.5)
Year 4	41 (10.8)	26 (6.9)	n/a	7 (1.9)
Year 5	49 (13.0)	21 (5.6)	n/a	8 (2.1)
Knowledge on Risk Factors				
Eating fast foods	279 (73.8)	97 (25.7)	n/a	2 (0.5)
Smoking tobacco	228 (60.3)	145 (38.4)	n/a	5 (1.3)
Physical inactivity	294 (77.8)	78 (20.6)	n/a	6 (1.6)
Drinking alcohol	315 (83.3)	61 (16.1)	n/a	2 (0.5)
Knowledge on Prevention				
Avoiding sweet foods	194 (51.3)	184 (48.7)	n/a	n/a
Diet modification	266 (70.4)	112 (29.6)	n/a	n/a
Regular exercise	175 (46.2)	203 (53.8)	n/a	n/a

Abbreviations: DM, diabetes mellitus; n/a, not applicable.

Among the 378 participants, the majority (n=323, 85.4%) reported that diabetes mellitus is a preventable disease, under half (n=44, 11.6%) disagreed, and a minority (n=11, 2.9%) did not know whether or not diabetes mellitus is a preventable disease. According to the respondents, the top 3 ways to prevent diabetes was by avoiding sweet foods (51.3%), diet modification (70.4%), and regular exercise (46.2%).

Attitudes of Participants Towards Diabetes Mellitus

Almost half (48.1%) of the study participants felt that personal efforts would not help control one’s risk of getting diabetes. In general, our results showed that minority (8.9%) of the study participants felt that if one was to get diabetes

mellitus, there was not much one could do about it, while the majority (90.8%) thought that one could do something to prevent getting the disease. This correlated with majority (89.6%) of positive responses to the statement that ‘people who make a good effort are less likely to get diabetes’ (Table 3).

Perceived Risk Related to Diabetes Mellitus

A majority (85.1%) of the participants disagreed with the statement that they did not have to worry about diabetes until midway into their fifth decade of life (age 45), while only 14.8% thought otherwise. Nearly half (47.9%) of the 378 students reported they were less likely to get diabetes mellitus compared to their age mates. A majority (84.1%)

TABLE 3. Participants’ Attitudes and Perceived Risk Related to Diabetes Mellitus (N=378)

Question	Strongly agree n (%)	Agree n (%)	Disagree n (%)	Strongly disagree n (%)	Not answered n (%)
Attitude Related to Diabetes Mellitus					
I have little control over risks to my health	28 (7.4)	55 (14.6)	152 (40.2)	140 (37.0)	3 (0.79)
If I am going to get DM, there is not much I can do about it	10 (2.6)	24 (6.3)	150 (39.7)	193 (51.1)	1 (0.26)
My personal efforts will help control my risks of getting DM	9 (2.4)	182 (48.1)	14 (3.7)	168 (44.4)	5 (1.32)
People who make a good effort to control the risks are much less likely to get DM	140 (37.0)	199 (52.6)	22 (5.8)	15 (4.0)	2 (0.53)
Perceived Risk Related to Diabetes Mellitus					
Compared to my age mates, I am less likely to get DM	60 (15.9)	121 (32.0)	143 (37.8)	47 (12.4)	7 (1.85)
I should not worry about DM until I am 45 years old	20 (5.3)	36 (9.5)	140 (37.0)	182 (48.1)	0 (0)
DM could be a big threat to my health	188 (49.7)	130 (34.4)	28 (7.4)	32 (8.5)	0 (0)

Abbreviations: DM, diabetes mellitus.

reported that the disease could be a big threat to their health, while the remaining 15.9% said indicated otherwise (Table 3).

DISCUSSION

Almost all (99.2%) of the participants in our study knew about diabetes mellitus. These findings are different from what was reported among medical students in Saudi Arabia, where knowledge of the prevalence of the disease among medical students was less than 30%.¹² A study in Libya, however, reported high knowledge levels (76.7%) related to diabetes mellitus among the medical students.¹³ The Libya study, however, sampled final year medical students whereas our study population sampled students across all the years. Students in their final year are more likely to have more knowledge since they are on their final path to medical practice. The high level of knowledge among our study participants may be due to the compulsory community attachment part of the medical student curriculum that mandates students work in communities and are expected to interact with individuals in the community and evaluate the key health problems within those communities. This

practical experience our study participants receive is important because in developing countries general doctors play a very important role in chronic disease management. This is also important because the prevalence of diabetes mellitus is increasing, with sub-Saharan Africa reported to be one of the regions with the fastest growing rates of diabetes mellitus in the world.^{2,3} Our findings were comparable across almost all the medical schools in Uganda, which may be attributed to the similarity of the medical training curriculum in the different medical schools.¹⁴ In our study, there were more male participants compared to females, reflecting current enrolments in medical school. We did not assess differences in the levels of knowledge between the genders since they attend similar lectures; however, this has been noted as one of the limitations of our study.

The majority (86%) of students had good knowledge about the signs and symptoms of diabetes. Participants knew that diabetes mellitus may present as excessive hunger (82.5%), blurred vision (74.6%), and frequent urination (86.8%), respectively. In contrast to a study by Khan et al. among university students in Ajamn, United Arab Emirates (UAE), students reported excessive eating (36.0%), blurred vision (39.0%), and excessive urination (58.0%) as

symptoms of diabetes mellitus.¹⁵ More than 70% of participants reported hunger, blurred vision, fatigue, and frequent urination as symptoms of diabetes mellitus. In contrast, Kulkarni et al. reported that medical students were able to identify the most common symptoms of diabetes mellitus, such as increasing thirst (14%), increasing hunger (8%), and weight loss (6%), although the knowledge levels were also low (6%–34%).¹⁶ The differences in knowledge levels may be due to differences in prevalence of diabetes mellitus in the different regions, differences in school curricula in the different regions, or differences in health agenda. Seven participants in our study demonstrated absolute ignorance about the signs and symptoms of diabetes mellitus: 5 were from Year 1, 1 from Year 2, and 1 from Year 3. This shows that the level of knowledge probably increases as one proceeds through the years in medical school.

The majority (83.1%) of the respondents reported that genetic factors were responsible for diabetes mellitus causation, a figure higher than the 60% found by Poornima et al.¹⁷ In a study by Shu Hui et al., a majority of patients were not aware of the causes of diabetes mellitus.¹⁸ This underscores the importance of health workers having proper knowledge of the causes of diabetes mellitus so they can educate patients accordingly. However, the study by Khan et al. also reported that university students were able to identify genetic factors as risk factors for diabetes mellitus.¹⁵ Because of the increased prevalence of NCDs, it is likely that this high level of knowledge among students is due to health education campaigns about diabetes mellitus in the media.

Among the 378 respondents, the majority (n=323, 85.4%) reported that diabetes mellitus is a preventable disease, comparable to a study among university students in Ajman, UAE, where 74% of the students reported that diabetes can be prevented or delayed.¹⁵

Whereas diet modification was reported by the majority (70.4%) of students as a method of diabetes mellitus prevention, avoiding sweet foods was not as highly regarded, with only 51.3% of respondents reporting it as a method of preventing the disease. The respondents further reported that consumption of fast food (73.8%) and smoking (60.3%) would predispose to diabetes mellitus, while a reduction in alcohol (83.3%) would help prevent diabetes mellitus.

Physical activity is a factor that has been reported to reduce mortality and also lead to improvement among patients with diabetes mellitus. Therefore, it is surprising that up to 53.8% of our respondents reported that regular exercise is not important in diabetes mellitus control; this is despite the report that revealed that inactivity predisposes to diabetes mellitus. Similarly, Shu Hui et al. reported in their study that less than 45% of diabetic patients perform regular exercise to improve diabetes mellitus control.¹⁸ Not surprising, a study by Kulkarni et al. among college

students reported that only 12% of students reported sedentary lifestyle as a risk factor for diabetes mellitus.¹⁶

About two-thirds (65.6%) of our respondents did not have a family member with diabetes, regardless of the high knowledge levels reported. A family history of diabetes has been reported to influence the students' level of knowledge and perception of diabetes.¹⁹ This is probably because they will be directly involved in patient care, which also involves family health education.

A majority (85.1%) of the respondents disagreed with the statement that they did not have to worry about diabetes until midway the fifth decade of life (age 45) and majority felt there was much one could do to prevent oneself from getting diabetes mellitus. A large percentage (84.1%) reported that diabetes mellitus could be a big threat to their health. Half (50.2%) of the students perceived themselves at more risk of getting diabetes mellitus than their age mates, however, almost the same percentage (50.5%) felt that personal efforts would help control one's risk of getting diabetes. One of the biggest weaknesses of our study was that we did not assess the students' own practices towards control of diabetes mellitus; this would have illustrated if those who perceive themselves at risk and believe that their personal efforts can help prevent them from contracting diabetes mellitus are actually doing something about it.

Another limitation noted was not ascertaining which students were also patients living with diabetes and comparing this to their level of knowledge of the disease.

CONCLUSION

Majority of respondents exhibited high levels of knowledge of the causes and symptoms of diabetes mellitus. The respondents also exhibited a positive attitude towards prevention of the disease. A majority further perceived themselves at risk of getting diabetes. However, this study revealed some gaps in the knowledge of medical students concerning the prevention of diabetes mellitus that can be appropriately used in designing the curriculum for the different medical schools. The study also lacks data to address the practices of the students as regards to their perceived risk. To that end, we recommend future studies fully evaluate the practices of university students related to diabetes mellitus.

Acknowledgements: We would like to extend our sincere gratitude to Medical Education for Services to All Ugandans (MESAU) – Medical Education Partnership Initiative (MEPI) (MESAU-MEPI) for giving us financial support. The project described was supported by Award Number 1R24TW008886 (MESAU-MEPI Programmatic Award) from the Fogarty International Center. The content is solely the responsibility of the authors and does not necessarily represent the official views of the Fogarty International Center or the National Institutes of Health.

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- Peer Reviewed**
- Competing Interests:** None declared.
- Received:** 31 Oct 2016; **Accepted:** 31 Jul 2017
- Cite this article as:** Kharono B, Nabisere R, Kiddu Persis N, Nakakeeto J, Openy A, Bakeera Kitaka S. Knowledge, Attitudes, and Perceived Risks Related to Diabetes Mellitus Among University Students in Uganda: A Cross-Sectional Study. *East African Health Res J.* 2017;1(2):105-112. <https://doi.org/10.24248/EAHRJ-D-16-00371>
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Implementing a Global Health Qualitative Research Study: Experiences of a Project Coordinator in Uganda

Jasmine Kastner,^a Cecilia Milford,^b Cecilia Akatukwasa,^c Annet Kembabazi,^c Jenni Smit^b

^aMcGill University Health Centre, Research Institute, Montreal, Canada; ^bUniversity of the Witwatersrand, Faculty of Health Sciences, Durban, South Africa; ^cMbarara University of Science and Technology, Mbarara, Uganda
Correspondence to Jasmine Kastner (jasminekastner@gmail.com).

ABSTRACT

Qualitative research in global health requires substantial operational and logistical support during both the implementation phase and day-to-day operations. However, little to no published work shares the experiences of international qualitative research teams. Yet, without a strong project foundation and attention to everyday details, studies can begin without appropriate guidance and, as a result, poor quality data may be generated. This paper presents a detailed account of a project coordinator's experience implementing 4 qualitative HIV and reproductive health studies in Uganda between 2012 and 2014, reflecting on our research team's practices and lessons learnt, and provides recommendations for successful project implementation. The aim of this paper is to help new global health qualitative project coordinators, and international teams more generally, by detailing 6 coordination tasks: hiring, training, team communication, organization of study documents, data collection and storage, and research ethics. To avoid repeat learning of basic, yet important, logistical steps by each new qualitative research project coordinator, this paper can help coordinators think about how to organize their work in order to prepare for both planned and unplanned challenges that have been encountered by others. Sharing operational and logistical experiences and expertise can benefit the global health community and help future studies run more efficiently.

INTRODUCTION

While quantitative research still dominates global health, the use of qualitative research has increased over the past decade. The HIV/AIDS epidemic has been at the forefront of combining epidemiologic and social science research to address the complex mix of social, cultural, political, and economic factors related to the virus and new treatment methods.¹⁻¹⁰ Powerful on its own, qualitative research has long been a crucial foundation for biomedical intervention programs, cohort studies, and clinical trials, as the success of these larger studies is context dependent.^{2,11-15} Qualitative research is essential to help identify and address the multiple realities of clinic patients and participants prior to planning and implementing interventions, programs, or large quantitative studies.

International qualitative research studies require significant operational and logistical support on the ground during both the initial set up of the research infrastructure and day-to-day operations. At times, the project coordinator position may be filled by a young professional

beginning a career in global health research. Extensive studies have focused on building research capacity and optimizing the execution of a range of specific quantitative research methods in a successful and efficient manner.¹⁶⁻¹⁹ However, little information has been published on the key components of research logistics and the day-to-day operations of running a research study in the field, especially regarding qualitative research studies. The sharing of on-the-ground experiences and best practices is necessary to enhance and ensure rigour in qualitative research.

The global health community habitually emphasizes their goal to improve health for all and move towards equity for people worldwide.²⁰ Qualitative research is needed to provide in-depth and contextual data about socio-behavioural issues that impact health and decision-making and to better understand new methods of improving health. Yet, little to no information is shared within the global health community about the important steps needed to conduct qualitative research during implementation of these studies. Without this knowledge, individual research groups may begin their studies

without guidance or suggestions from experience gained or lessons learnt from previous studies. This can result in ad hoc qualitative research practices, which may undermine the importance of rigour in qualitative research. We believe that the research community should collectively grow from shared experiences and conversations about the common steps required to achieve quality qualitative data. For example, the methods section in most journal publications is intended to provide a replicable description of data collection and analysis procedures. Yet, the preparatory steps leading up to data collection; the logistics required to achieve and maintain high data quality, research ethics of the highest standards, and team formation, training, and participation; and overall study operations are rarely documented.

In this paper, we present a detailed account of our international team's experiences implementing 4 qualitative studies in Uganda between 2012 and 2014. We reflect on our experiences and lessons learnt, and provide recommendations for successful qualitative project implementation. This paper focuses on the 6 coordination tasks that will help guide new global health qualitative research project coordinators as they implement research studies in the field.

Our intention is to share our team's experience in order to provide first-time qualitative research coordinators with an organizational foundation that could facilitate the running of future studies smoothly. We believe that this paper will help project coordinators organize their work by being aware of and planning for planned and unplanned challenges that have been encountered before. Encouraging research teams to share their operational and logistical expertise will benefit the global health community as they build on each other's experiences. In turn, this will help researchers avoid repeat learning of the same important steps and anticipate challenges when implementing a new study.

BACKGROUND

Our qualitative studies were designed as a follow up to quantitative study results^{21,22} and as a first step towards implementing sustainable HIV and reproductive health programs. All 4 of our global health qualitative studies were focused on HIV, reproductive health, and access to care, and were conducted in Mbarara, Uganda. Uganda has one of the highest total fertility rates globally, estimated at 6 children per woman.²³ HIV prevalence among adults (aged 15–49 years) is estimated at 7.3%, with higher prevalence among women (8.3%) compared with men (6.1%).²⁴ Our studies focused on in-depth interviews with: 1) serodiscordant couples regarding their pregnancy plans;²⁵ 2) health care workers about their views and knowledge of reproductive health care for HIV patients;²⁶ 3) men living with HIV about their practices and motivations around disclosure and family planning practices and; 4) recently pregnant women living with HIV and mental health care workers about postpartum

depression.^{27,28} These qualitative studies were developed from an ongoing reproductive health study within the Uganda Antiretroviral Rural Treatment Outcomes (UARTO) cohort study²¹ in Mbarara, and were all separately approved under national and university research ethics boards. All participants living with HIV and their serodiscordant partners were recruited from the UARTO cohort. Health care workers were recruited by contacting clinics within the district of Mbarara.

For the purposes of this paper, the qualitative research methods used were in-depth interviews and field notes. The goal of our studies was to collect individual perspectives and lived experiences. Given the sensitivity of our research topics, our team collectively chose in-depth interviews as the most confidential and secure method for participants to share their stories. Interview guides and other supplementary information about these 4 studies can be found in the referenced publications.

Our Ugandan studies were international collaborations, which added to the complexity of the research process. Principal investigators were located in Uganda, Canada, South Africa, and the United States. The Canadian project coordinator was based in Mbarara for the duration of the studies and was supported by 3 Ugandan research assistants. The majority of team members had previous experiences implementing studies in Mbarara, Uganda.

In the following sections, we share our team's experience by outlining 6 important tasks for implementing qualitative research studies in global health with a multi-national team. In each section, we break down our experience into implementation, lessons learnt, and recommendations for best practice (Figure 1).

BUILDING A RESEARCH TEAM

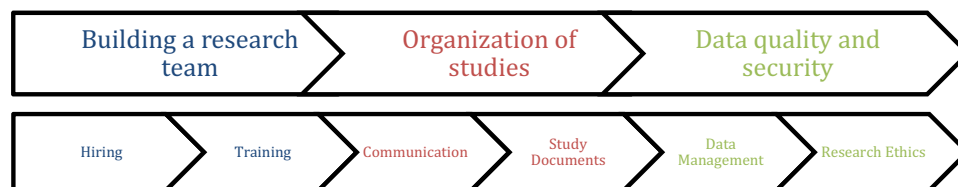
Hiring Field Staff

Implementation

A research job advertisement in Uganda, where unemployment rates for the under-30 working population is 35%,²⁹ receives many applicants. Our interview committee consisted of the Ugandan principal investigator, the project coordinator, the human resource manager, and a second project coordinator. Having a diverse interview committee helped us conduct thorough interviews with the candidates, and rank each candidate's interview responses and work experience.

Our interview committee was equipped with a list of questions for all candidates that covered qualitative research experience, general work experience, work ethic, the understanding and ability to implement the principles of research ethics, personal expectations, and personal interest in the research topic and method. We also explored their personal values and level of comfort talking about the sexual and reproductive health issues we were researching. These questions helped us to assess if a candidate would be a good match for our research team.

FIGURE 1. Experience and Advice on Key Components to Study Logistics



At the end of each interview, the candidate was asked to conduct a mock interview with 1 person on our interview committee. This mock interview assessed the candidate’s comfort conducting an interview focused on HIV and reproductive health, and his/her fluency in the language in which the study’s interviews were to be conducted. The combination of the prepared questions and the mock interview helped the interview committee evaluate a best-fit candidate for the research team.

Lessons Learnt

Although conducting a mock interview with each candidate was time consuming, this method of assessing the applicant’s qualifications was indispensable. Through these interviews, we learnt that while not all eligible candidates may have advanced degrees or experience in qualitative research, consideration should also be given to candidates who show natural interview skills and a personality that is compatible with the research team. During fieldwork, we also learnt that hiring research assistants with similar demographics as the potential participants – such as age, sex, tribe/ethnic group, and community – helps make participants feel comfortable to open up and share their stories during interviews. Our team also took on a student trainee who benefited from the training and was later hired as a full-time research assistant.

Recommendations for Best Practice

We recommend including bi-annual evaluations in an employee’s contract. These coordinator evaluations ensure formal feedback is prepared about the employee for both the principal investigators and employee her/himself. The bi-annual evaluation should be formally written and discussed in a meeting between the project coordinator and research assistant, as well as by a conference call with the international principal investigators. The evaluation should focus on the research assistant’s successes and areas for improvement. The formal feedback mechanism is appreciated and seen as a good time to discuss career goals shared during the hiring process and a way to guide professional development. It also serves as a platform to address any

problems and areas to continue working on. This process enables the project coordinator to facilitate career advancement when possible; an essential part to increasing research capacity at the field site.

Training Implementation

Over the course of a study, staff training is a continuous process, with the bulk of training concentrated at the beginning of a project. While the initial training for each study was time consuming, it contributed to the study’s success. Our project coordinator spent 2 weeks of full-time training to familiarise the team – 3 new research assistants and 1 student – on the study goals and methods. The initial training focused on the differences between qualitative and quantitative research, background of qualitative research, different interview techniques, body language during interviews, research ethics and maintaining confidentiality (including informed consent and interview protocol), conducting practice interviews, standard operating procedures, and an introduction to data analysis. Training information was provided to research assistants through oral presentations, PowerPoint slides, research articles, training articles, and fact sheets. Even though some research assistants had previous qualitative research experience, all team members agreed that the basic training helped them achieve a common skill level, set values, and purpose to the studies. It was also a useful way for the project coordinator and the research assistants to get to know each other.

Once the initial training on general qualitative research methods was complete, the project coordinator built on the qualitative skills recently learnt by introducing the protocol of the first study. This enabled all staff to understand the purpose of the study, previous work that had been completed in the area of research, and approach to be used to conduct the study. We then moved on to review the interview guide and conduct mock interviews with the research assistants. The mock interviews provided research assistants an opportunity to become comfortable with the study interview guide and understand the purpose of each question, while practicing their qualitative interview skills.

Lessons Learnt

Consistent with previous research,^{30,31} we found that training in a small group created a productive learning environment. Peer review and feedback were also an essential part of our training method. These activities gave Ugandan research assistants the opportunity to learn from each other and provide invaluable cultural insight and suggestions for changes to the study documents. These suggestions improved the interview guide and made for more successful interviews, something that would have been lost to non-local staff.

Practising with the study's interview guide also provided the project coordinator an opportunity to understand research assistants' level of knowledge about the health background of the study – in our case, HIV, reproductive health, and mental health. We learnt that if research assistants were not comfortable explaining the study purpose or responding to basic questions about the research topic to peers, the actual study interviews lacked the in-depth content desired. Moreover, understanding the purpose of why each question is included in the interview guide was imperative, as was allowing the research assistant to expand or rephrase questions during an interview so that a participant could share their story. Secondary training was conducted by the project coordinator whenever a research assistant struggled to explain the purpose of the study or a particular question. When necessary, the purpose and background of the study was reviewed. Our team prepared multiple short trainings on HIV, family planning, and mental health over the span of the projects. These trainings were critical to the success of our qualitative studies since they instilled confidence in the research assistants when conducting the interviews and facilitated understanding of when and where to probe deeper on participant responses during interviews.

Although an intense initial training program did lead to an initial time lag in data collection, the time dedicated to training and preparation was more than compensated by structured data collection and good quality data. Training research assistants on interview techniques and protocol should not be limited to the beginning of the study, it is a process that should continue throughout the duration of a study. We learnt that feedback from the project coordinator and principal investigators after the first few interviews of a study helped highlight any persistent misunderstandings about certain questions in the interview questionnaire. Providing constructive criticism was often welcomed as a way to improve personal interview skills and the overall quality of the study. We also learnt that these feedback conversations created a comfortable space for research assistants to express any struggles they may have with asking certain questions or with the flow of the interview guide.

Recommendations for Best Practice

We recommend that feedback to research assistants should include a review of the purpose of the study, relevance of

each interview question, research ethics protocol, and suggestions on how to improve probing during participant interviews. The training and review processes listed in the above section help to ensure that research and ethics protocols are maintained while improving the quality of the study data collected. We conducted regular protocol reviews with the research assistants in order to keep the team up to date. A training and review process can create a positive feedback loop that benefits both research assistants and the study findings. We noted that with better information, the data improved, and, subsequently, the confidence and skills of the research assistants also improved.

Feedback from research assistants during the training periods and initial data collection can help improve the quality of the study through small modifications to the interview guide and study protocol, ensuring that studies are optimized for the respective cultural framework(s). Such small modifications may include revising the wording of interview questions to address social and cultural norms, which may require research ethics amendments, and enable better flow of the interview guide, more thorough responses from participants, more appropriate translation of study documents, and improvement of data quality. We submitted all interview guide modifications to our respective research ethics board (REB). Uniformity across all interviews remained since participant interviews began only after the initial training period – where modification to study documents were made – and after REB amendments were approved.

ORGANIZATION OF STUDIES

Communication

Implementation

Team communication is important for any study. Since our qualitative research team was small (less than 10 people in total), the project coordinator, 3 research assistants, and 5 principal investigators scheduled regular meetings to discuss how the interviews were progressing and to anticipate any possible upcoming difficulties. It was essential for our local team to remain in daily contact in order to make sure the interviews were up to our standards and discuss any logistical problems. Day-to-day communication was conducted with field site staff – 1 project coordinator, 3 research assistants, and 2 principal investigators – both in person and via email or Skype when including the 3 international principal investigators. Beyond the local team meetings, our project coordinator and international principal investigators met weekly over Skype to discuss the progress of the studies.

Lessons Learnt

Commitment to scheduling and preparing team meetings required flexibility since we were based across multiple time zones. Coordinating meetings between Uganda, Canada, South Africa, and the United States required some staff to participate outside normal working hours, meeting agendas

to be circulated early, and access to well-functioning communication networks. Occasionally, online meetings had to be re-scheduled or changed to email or phone conversations when the internet in Uganda was not reliable. It was the role of the project coordinator to organize team meetings and ensure that all relevant documents were circulated beforehand.

Having regular team meetings – both via telecommunication and in-person – helps integrate all research members into the study and resolve potential challenges efficiently. Team meetings also provide accountability checkpoints, since all members may be assigned work tasks. Team member progression on these tasks can be recorded in the meeting notes, circulated to all, and discussed at the start of the following meeting. In our project, knowing that all team members were working on a study task, helped boost individual work levels, as everyone understood their role in the team and responsibility in the team effort.

Recommendations for Best Practice

As a study progresses, it is beneficial to encourage research assistants or younger team members to lead team calls or meetings and provide feedback on how fieldwork is progressing. The importance of research assistants leading calls or meetings is aligned with the bilateral training and review process (see the Study Documents section). In a small qualitative research team, these steps empower research assistants or other team members to help develop the study rather than just follow assigned tasks. Active participation in study calls or meetings also means that all study members have an understanding of the larger framework and purposes of the study – that they are not just performing activities in isolation for the bigger project. In our study, interactive communication – through sending emails to principal investigators, sharing opinions during meetings, and asking for clarification – enabled research assistants to develop a sense of ownership in the work they were doing while promoting capacity building.

Finally, we recommend informal conversations amongst onsite staff about the progress of interviews and debriefs after each interview. Such conversations are also opportunities to provide reminders about training points, help research assistants improve interviews, and debrief about difficult interviews. In our study, these conversations were initiated by the project coordinator, with respective research assistants, and facilitated the operation of each study.

Study Documents

Implementation

Beyond study protocols, research ethics documents, interview guides, and other general study documents, we found 3 sets of documents to be essential to the success of our qualitative research studies. These 3 document sets included: standard operating procedures (SOPs), interview recruitment scripts, and memorandums of understanding (MOU).

While commonly used for quantitative studies, these documents are not often required for qualitative studies. However, we believe they should be part of project implementation. The documents were developed by the project coordinator during the initial phase of our studies, outlining all study procedures in detail. All of these documents ensured that our team members were following the same guidelines and study protocols, and facilitated smooth study operations, and could be referenced when needed.

It was important to design protocols and SOPs that described all steps of our studies. These documents outlined operational procedures and were the primary reference source for future interest in replicating our studies. More importantly, the SOPs were used throughout each of our studies to set a general standard for all logistics. SOPs helped reinforce that study procedures were implemented in a uniform fashion, and ensure data collection practices met our high standards.

The SOPs were a resource for all study members. They described in detail how to schedule an interview, what documents and equipment were required for each interview, what was considered best interview etiquette, how to ensure confidentiality for participants, what transcription procedures should be used, where to record and store all data securely after each interview, where to record study expenses, and where all documents and supplies should be kept. All study procedures were written by the project coordinator and circulated to team members for review and input before being finalized. SOPs were circulated to all onsite team members in print and uploaded to the study cloud storage space for all to access as needed (more on this in the Data Quality and Security-Implementation section).

Lessons Learnt

After reviewing early participant recruitment notes and questions posed by participants during the interview, we learnt that it was important for research assistants to have a clear and consistent script to introduce the study to eligible potential participants. These scripts were available in the local language and could be used for in-person or phone conversations.

The short scripts were used to guide the research assistants during participant recruitment and were useful tools to guarantee uniform recruitment and a clear presentation of the study purpose and details to eligible participants. They also ensured that all potential participants were aware that the study would include an interview; that they would be compensated for their time; and that we would call the day before their scheduled interview day to confirm their participation. We also learnt that this last point was important for planning our daily work schedules and minimizing unnecessary money spent in the study budget for transport.

SOPs and recruitment scripts were particularly useful during the early recruitment phase when research assistants were still getting used to the study. These documents helped

to guarantee that all important details were shared with eligible participants. As the studies progressed, SOPs and scripts were phased out and only used when necessary. All study protocols and scripts were reviewed and shared with team members.

Recommendations for Best Practice

Finally, when principal investigators are located across many countries, we recommend that an MOU between each investigator be written. An MOU describes the expectations and specific roles of each principal investigator during each phase of the study. It is ideal to create and circulate this document during the early planning stage of a study. This ensures that all principal investigators agree on their respective workload during the study design, implementation, analysis, write-up, and dissemination of the study. Once agreed upon, an MOU is a contract to help ensure that the study runs smoothly and each designated principal investigator completes their assigned section of the study on time.

DATA QUALITY AND SECURITY

Data Management

Implementation

This section will explain the procedure we followed after each interview was conducted. Although qualitative research differs from clinical trials and quantitative research, research ethics guidelines and good clinical practices must be followed to protect participants and ensure good-quality research. Our team made the decision that only 1 interview per research assistant could be completed per day. This assured uniformity across all interviews. The rest of the day was spent preparing for the interview, completing field notes after the interview, meeting with the coordinator to discuss how the interview went, and beginning the transcription process.

The project coordinator prioritized safe storage of all interview data. Documenting demographic data and creating summaries after each interview, rather than logging data at the end of the study, helped our team with discussions about the progress of our studies and the identification of interesting findings. This helped us to prepare for conference paper submissions, write progress reports, and present informal findings to our research collaborators. Furthermore, by updating study data and information after each interview, we were able to identify any mistakes and resolve any queries with the participant, timely and with accuracy.

After each interview, the research assistant who conducted the interview added information to 3 ongoing spreadsheets shared with all team members: demographic data collected at the start of the interview; interview details, such as location of the interview and expenses incurred; and short interview summaries. Collecting a brief summary of each interview in a single document was useful during the

analysis phase of the study, as it provided a quick overview of the main points of an interview and saved time that would have been spent reading lengthy transcripts.

The next step in the data collection process was for the research assistant to transcribe and translate the new interview from the local language to English. This was done using Express Scribe software and following the relevant SOP. Once complete, a different research assistant reviewed the interview transcript while listening to the audio. This process provided an opportunity to clarify translation errors, catch phrases that were missed during transcription, and correct misspellings. The final review was completed by the project coordinator and, sometimes, a principal investigator to clarify any cultural concepts or questions in the transcript.

Lessons Learnt

It is important to note that transcription and translation activities are time consuming for all staff. One hour of audio can take from 6 to 8 hours to be translated and transcribed. The transcription review by a separate research assistant typically takes 2 to 3 times the length of the interview, as does the final review. Additional time for feedback from each interview must also be added, especially during the early stages of a study. Given this, an average of 1 to 2 interviews per week per research assistant was our norm, depending on the length of the interview. Our studies did not conduct any focus groups, but it is important to note that the transcription of focus groups often takes longer than one-on-one interviews.

Research assistants were encouraged to add field notes at the end of the interview transcription. Writing field notes is standard protocol in qualitative research³²⁻³⁶ and provides an opportunity for the research assistant to explain how the participant was acting, what body language was used, and any additional information not available from the audio recording. Along with the short interview summaries in the shared spreadsheet, we found that field notes often helped our team understand the interviews better since they summarized the interview and included additional comments from the research assistant who conducted the interview.

All study documents were secured with standardized file names, for identification ease; audio files and password-protected transcripts and spreadsheets were stored on an encrypted and password-protected virtual cloud. This secure virtual cloud allowed for real-time access to all authorized team members. We learnt that having 1 person responsible for storing and organizing study documents helps to avoid confusion. Our project coordinator was in charge of keeping all study documents up-to-date and all study folders organized on the virtual cloud and in hardcopy format. This role included ensuring that the most recent SOPs, study budgets, progress reports, interview guides, transcripts, summaries, and demographic data were uploaded online. Signed consent forms were stored as hardcopy documents only, and all

hardcopy study documents and equipment were stored in a key-locked filing cabinet in a secure research office.

Recommendations for Best Practice

Our project coordinator's role was to ensure that all interview information – such as an identification number associated with the person interviewed, location and time of interview, and transport refunded – was entered into the online spreadsheet timely and accurately; to conduct a final review of all transcripts; and to ensure demographic data for each participant was up-to-date. We recommend that a designated staff member be responsible for removing the audio file from the voice recorder and storing it in a protected and safe backup location. Once the interview transcript has been reviewed and queries clarified, the electronic transcript can be stored with its respective audio file for future reference.

Following good clinical practice guidelines, labelling and organizing hard copy storage of all research ethics documents will save time when writing REB renewals or searching for study data. We recommend assigning separate files for storing signed informed consent documents, research ethics documents, SOPs, scripts, and interview guides. Organizing hardcopy files is useful when documents have to be shared with other research collaboration team members, when there is need to consult original participant documents, or when it is necessary to prepare for an audit.

Research Ethics

Implementation

While research ethics falls under the 'data quality and security' heading, this theme cross-cuts all sections of this paper due to its importance throughout the entire research process: from preparing study documents, screening participants, recruitment, consent, interviewing, and data collection and analysis, to the dissemination of findings. The principal purpose of research ethics is to ensure the participant's rights are not violated in any way, and that confidentiality is maintained. In qualitative research studies, research assistants often conduct interviews about personal and sensitive topics – in our case, HIV, sexual practices, depression, partner relationships, and pregnancy. All of our interviewers were trained – through online and in-person ethics and good clinical practice workshops – in maintaining research ethics and protecting the rights of the participants that may arise in any interview situation. Training was also provided to ensure the research assistants conducting interviews were comfortable pausing or stopping an interview if a participant was uncomfortable, not ready to continue, and distressed; if they needed to be referred for counselling or treatment; or if someone interrupted the interview. Most interviews were conducted in a private room adjacent to the HIV clinic, with only a few participants electing to conduct the interview in the privacy of their own home.

Lessons Learnt

We learnt that participants in our studies sometimes assumed that the research assistant conducting the interview was a health care provider and sought advice from them. This occurred despite the fact that during study introduction and the consent process research assistants stated that they were not providers. The project coordinator provided research assistants with training on how to avoid feeling as though providing health advice may be the only way to move forward with the interview. While it was necessary for research assistants to understand the health background of the study in order to conduct thorough interviews, it was equally as important for the research assistant to understand that their role was to conduct a thorough interview and not to provide health advice they were not professionally trained to give.

After the first encounter of this situation, the project coordinator designed a health care referral form and SOP for interviewers to use if their participants asked for health advice. The SOP included a script for explaining that the interviewers are not health care professionals but could refer the participant to a health care professional after the interview. The referral form included the name and phone number of health care professionals from HIV and mental health clinics who had volunteered to speak with any participants if they had further health questions.

Recommendations for Best Practice

Beyond maintaining research ethics before, during, and after interviews, it is the role of a project coordinator to apply for research ethics renewal and request approval for amendments to all studies. Missing an REB deadline can force a study to pause data collection while waiting for REB renewal approval, resulting in a loss of time, money, and the pace of the study. Having a multi-national collaboration required our project coordinator to keep track of research ethics approvals and renewal dates for 4 different studies across 3 different countries. We recommend that the project coordinator create 1 document and 1 calendar that lists all research ethics committee approvals and expiry dates to ensure timely research ethics renewal applications amidst other operational tasks.

Since our research team was based within a larger research collaboration, we received help with submitting our application packages to the 4 institutions for which our studies required REB approval. The advice and help received from our colleagues was invaluable in navigating international research board applications, amendments, and renewals. We recommend that first-time project coordinators seek advice from colleagues with experience. Knowledgeable colleagues can share research ethics cover letters and application forms, and provide insightful tips for submitting REB application packages correctly. They can also advise on the estimated time for study feedback on application submissions, thus helping with appropriate study planning.

DISCUSSION

Interest in qualitative research in global health is growing. Over the course of our studies presented in this paper, many of our research collaboration team members were excited to learn about our work and expressed interest in learning more about the qualitative research methods used in our studies and the lessons shared in this paper. The points presented are a combination of lessons learnt and recommendations for project coordinators to manage operations and ensure their future projects move forward smoothly while assuring data quality. These points are useful references for both first-time and experienced qualitative research project coordinators in global health. Any new international qualitative research team member can benefit from learning from the shared experiences of other researchers (Figure 2).

While each of the key points are presented separately, they are interrelated and build on each other. Beyond what is shared in this paper, we recommend that a new project coordinator seek out a mentor at the field site to provide advice about the research collaboration – a group of researchers from different organizations working together in a similar field of research, sharing data and ideas – including expected timelines to complete requisite responsibilities, how REB application(s) should be prepared, who to contact for help with various work tasks, samples of previous work, and other general support. Collaborating with colleagues and peers is also a useful way to learn about the most efficient techniques

to manage various coordinator duties that are common across all studies at a field site. Collaborating with other project coordinators at the field site also promotes an open environment to ask for help when challenges arise.

Our team valued the assistance of our collaboration team members, the health clinic staff, and participants. Before beginning each study, we introduced the purpose of the study and methods used to clinic and research collaboration staff members so that all were aware of the work we were doing. We also disseminated our preliminary research findings at the end of the data collection phase to the collaboration and clinic staff in order to share the findings of our studies and maintain a good relationship. Both introductory and dissemination presentations are an important part of collaboration and conducting research. In doing so, a shared learning environment was created and our colleagues were helpful in moving the studies forward in order to learn more about HIV and reproductive health in the region.

In addition to the above, the Canadian project coordinator living in Uganda made it a priority to focus on equitable practices, which entailed open conversations about both work and social life, being open to sharing problems and asking for advice or help from colleagues, sharing a same small workspace with all team members, and attending research collaboration and clinic meetings. There were times where the international project coordinator was put into a position of power,^{37,38} yet when this occurred, navigating both international and local staff status was simplified through dis-

FIGURE 2. Summary of Key Points

Hiring	Training	Communication	Study Protocols	Data Management	Research Ethics
<ul style="list-style-type: none"> • Mock interview: comfort and language fluency • Research experience and natural skills • Personality match with other teammates 	<ul style="list-style-type: none"> • Thorough training in research ethics and good clinical practice • In-depth qualitative research training • Peer training and review, mock interviews • Subject of the research project • Continuous training and feedback over study 	<ul style="list-style-type: none"> • Team meetings, led by research assistants • Multi-national teams: Skype, phone, in person, virtual cloud, email • Separate project coordinator-principle investigator meetings • Informal meetings for field staff 	<ul style="list-style-type: none"> • SOPs for all important study tasks, reviewed by team • Scripts written to help research assistants with recruitment • MOUs between principle investigators at the start of the study • Prepare all study protocol documents before the study begins and modify where necessary over the course of the study 	<ul style="list-style-type: none"> • Responsibility of the project coordinator to keep study up-to-date • Interview audio and transcripts stored in two locations in a protected virtual folder • Continuous update of interview details • Secure virtual cloud storage 	<ul style="list-style-type: none"> • Sensitivity of qualitative research • Health care provider referral forms • Difference between conducting a well-informed interview and providing advice to participants • Keeping track of all research ethics committee applications and deadlines

Abbreviations: MOU, memorandum of understanding; SOP, statement of purpose

discussion about the tasks to be completed and the job roles of each person involved. Hard work and sharing the weight of tasks fairly was a way to avoid dogmatic views of international and local staff barriers within global health research. By constantly being aware of unspoken norms of power dynamics and privilege in global health research, the project coordinator focused on actively being part of the research and clinic teams by learning from, listening, and adapting to colleagues. Without the above-mentioned points in this discussion, our studies would not have been completed as efficiently or as successfully. Collaboration with other research teams, awareness of power dynamics, and sharing our study details with non-research colleagues created a productive work environment.

Finally, the operational and study logistic key points described in this paper are based exclusively on our qualitative research field site experience. Certain aspects of these points, like research ethics and study documents, can overlap with operational procedures of quantitative or mixed-method research. Yet, it is the combination of operational procedures across all 6 key components (Figure 2) that makes the operational tasks described in this paper unique to implementing a qualitative research study. It is also important to state that this paper focused on the data collection phase of qualitative research and did not delve into the rigorous process of data analysis to ensure results are interpreted correctly, or that they are reliable and valid.

CONCLUSION

Understanding your research collaboration's team dynamics, in combination with good interpersonal skills, is an important early step for any new project coordinator. Many of the key points explained in this paper can be completed through a 'learning-by-doing' process if project coordinators are provided with good mentors and peers for guidance. Reading about the experience and lessons learnt from previous studies is resourceful preparation for an upcoming project.

This paper hopes to promote communication within the global health qualitative research community. It is intended to help new project coordinators by detailing 6 key points that we believe help an international study team prepare for planned and unplanned challenges encountered by others in the past and assure data quality, while maintaining rigour in qualitative research.

Acknowledgements: We thank the principal investigators of the studies: Drs. Scholastic Ashaba, Francis Bajunirwe, Angela Kaida, and Lynn Matthews. A special thank you to both our mentors and peers within the Mbarara research collaboration who were open to sharing their experiences and knowledge, and without whom our studies would not have moved forward successfully.

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- Peer Reviewed**
- Competing Interests:** None declared.
- Received:** 28 Oct 2016; **Accepted:** 20 Jul 2017
- Cite this article as:** Kastner J, Milford C, Akatukwasa C, Kembabazi A, Smit J. Implementing a Global Health Qualitative Research Study: Experiences of a Project Coordinator in Uganda. *East African Health Res J*. 2017;1(2):113-122. <https://doi.org/10.24248/EAHRJ-D-16-00343>
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Health System Barriers to Provider-Initiated HIV Testing and Counselling Services for Infants and Children: A Qualitative Study From 2 Districts in Njombe, Tanzania

Gasto Frumence, Sirili Nathanaeli

Muhimbili University of Health and Allied Sciences, Dar es Salaam, Tanzania.
Correspondence to Gasto Frumence (frumencegasto@yahoo.co.uk; gfrumence@muhas.ac.tz).

ABSTRACT

Background: There is low utilisation of provider-initiated HIV testing and counselling (PITC) services for infants and children under 5 years old in many low- and middle-income countries including Tanzania. Studies have shown that various factors contribute to low use of PITC, includes the unavailability of the polymerase chain reaction (PCR) test and other specialised techniques for testing children less than 18 months old as well as the reluctance of some parents and caregivers to undertake HIV testing for their children because of the fear of stigma associated with HIV/AIDS. This study explored health system barriers at the district and community levels affecting the provision of PITC for infants and children under 5 in Tanzania using a case study of 2 districts.

Methods: A qualitative study was conducted in 1 urban and 1 rural district in the southern part of Tanzania. In-depth interviews, focus group discussions, and a desk review of documents were used to obtain the information. Respondents were purposively enrolled in the study and thematic analysis was used to generate findings.

Results: Provision of PITC services faces a number of district-level health system barriers, including lack of adequate health staff in health facilities both in number and skills, lack of adequate infrastructure, and erratic shortage of supplies. At the community level, community members' low understanding about the importance of PITC services as well as existing stigma associated with HIV/AIDS have constrained the provision of PITC services.

Conclusions: This study concludes that for effective implementation of PITC, the health system should strengthen health facilities through training of service providers on PITC, deploying adequately skilled health workers, supplying sufficient medicines and other supplies, and promoting health campaigns focused on educating community members about the importance of early HIV testing for infants and children under 5.

INTRODUCTION

Worldwide, approximately 36.7 million people were living with HIV/AIDS in 2016; of these, about 2.1 million were children and adolescents under 15 years of age.¹ In 2014 alone, it was estimated that 190,000 children were newly infected with HIV throughout Africa and about 3% of deaths in children under 5 years old were attributed to AIDS.² Most of these children were infected through vertical transmission, from mother to child, which accounts for more than 90% of deaths in children under 5 in sub-Saharan Africa.³ Despite various global and national efforts to scale up HIV prevention, care, and treatment, services have not been fully implemented for infants and

children who have been exposed or infected with HIV.⁴ The World Health Organization (WHO) recommends offering routine provider-initiated HIV testing and counselling (PITC) to persons attending health care facilities as a standard component of medical care.⁵ Early diagnosis of HIV infection in infants (0–12 months of age)⁶ and children under 5 is important for timely initiation of treatment with antiretroviral therapy.^{7,8}

Several studies have reported a number of barriers to testing children for HIV, including unavailability of the polymerase chain reaction (PCR) test and other specialised techniques in testing children less than 18 months old in many health facilities. The stigma associated with HIV/AIDS also contributes to the reluctance of some parents and caregivers to undertake HIV testing for their

children.⁹ Other barriers include poor linkages with prevention of mother-to-child transmission programmes and subsequent missed opportunities for testing during postnatal and child health care, challenges of providing virological testing required for early infant diagnosis (eg, RNA or DNA PCR), limited paediatric expertise amongst health care providers, and the prioritisation of adult treatment and subsequent lag in availability of paediatric antiretroviral therapy dosages.^{10–14}

In low- and middle-income countries, late HIV diagnosis and delayed care interventions result in high mortality rates amongst HIV-infected children.⁴ Tanzania adopted PITC in 2007 as one of the strategies for increasing access to HIV care in the country.¹⁵ PITC is a system whereby HIV testing and counselling are provided to all patients attending a health facility, regardless of the reason for their visit.⁴ Many countries in sub-Saharan Africa, including Tanzania, have developed and scaled up access to early infant HIV virological testing, particularly using dried blood spot testing.^{16–18} These blood spots are usually collected from health facilities and sent to referral hospitals' laboratories. However, this initiative faces several difficulties, such as the long delay between blood collection, the resulting large proportion of parents and caregivers who fail to return for the results, and the high number of HIV-infected infants who are lost to follow-up.³ In Tanzania, little is known about the barriers to early HIV diagnosis of infants and children under 5. Therefore, this study aimed to explore health system barriers at the district and community levels for early diagnosis of HIV in infants and children under 5 in Tanzania.

METHODS

Study Design

We used a cross-sectional exploratory study design to explore health system barriers at the district and community levels for HIV testing amongst infants and children and analysed results using a phenomenological approach. According to Lopez and Willis,¹⁹ a phenomenological approach seeks to describe the meanings embedded in the human experience and common life practices. Furthermore, this approach helps to explore the individual experiences as well as common characteristics and differences between groups of participants.

Study Setting

This study was carried out in Njombe Region located in the Southern Highland Zone. This zone was purposefully chosen because it has the highest HIV prevalence in the country, and, within it, the Njombe Region was selected due to its high HIV prevalence – 14.8% compared with the national HIV prevalence of 5%. Within the region, the Njombe and Makete districts were chosen to present the rural (Makete) and urban (Njombe) districts. The 2 districts also had high HIV prevalence in the region: 14.7% in Makete and 15.34% in Njombe District.²⁰

Study Population

The study population consisted of health service providers at regional and district hospitals and health centres where HIV testing for infants and children is done, parents and caregivers with infants and children under 5, and women who recently gave birth (in the past 42 days to 18 months).

Sampling Techniques and Recruiting Plans

We used the purposive sampling method to select 1 zone with high HIV prevalence amongst the 7 geographical zones. Within this zone, we purposively selected 1 region with high HIV prevalence, along with 2 districts with high HIV prevalence (1 urban and 1 rural). Within each district, the district hospital and 1 health centre from a randomly selected ward, amongst the wards with health centres, were included.

We also used purposive sampling to select health service providers, including the district hospital secretary, health facility in-charges, heads of care and treatment clinics, heads of laboratories, and heads of reproductive and child health units.

A convenience sampling was applied to select parents and caregivers with children under 5 and recently delivered mothers for conducting focus group discussions (FGDs). Recently delivered mothers included all women who had given birth in the previous 42 days (6 weeks) to 18 months. The choice for this post-puerperium period was based on cultural practices that hinder contacting mothers within the first 6 weeks after delivery. The upper limit was based on the fact that HIV antibody confirmatory testing in children is reliable after 18 months of age.²¹ Community health workers supported the process of identifying recently delivered mothers in their communities.

Data Collection Methods

A total of 18 face-to-face in-depth interviews were conducted with the district hospital secretary and all in-charges and heads of sections of selected health facilities. In addition, a total of 4 FGDs were conducted to gather information from families—2 FGDs with recently delivered mothers and 2 FGDs with parents and caregivers of infants and children under 5. Both in-depth interviews and FGDs aimed to explore district- and community-level health system barriers to HIV-PITC services for infants and children under 5. Data collection was stopped after 18 interviews and 4 FGDs after reaching the point of data saturation when no new themes could be found in the participants' statements. In-depth interviews and FGDs were conducted in health facility and village offices and lasted for 60 and 90 minutes, respectively. These offices provided adequate privacy without interference from people who were not participating in the study. The in-depth interview and FGD guides were developed, pre-tested, and revised before being used to collect data for this study.

A desk review focused on key strategic documents and guidelines on PITC services to clarify the themes that emerged from the in-depth interviews and FGDs and to

analyse the study findings in the context of Tanzania. This triangulation of data collection techniques – namely, the in-depth interviews with key informants, FGDs with recently delivered mothers, as well as desk reviews – helped to ensure the credibility of the study findings.²³

Research Team

The first (GF) and second (SN) authors and 2 research assistants were involved in collecting data for this study. GF and SN were responsible for conducting in-depth interviews and the 2 research assistants were responsible for conducting FGDs. It is worth noting that no relationship existed between the research team and study participants before their engagement in this study.

Quality Control and Data Management

Trained research assistants listened to the audio-recorded information every day to check the accuracy and sounds, and, where possible, to take corrective measures to rectify any missing information. Moreover, periodic team meetings with the principal investigators and research assistants were held to review progress.

Data Analysis

In this study, the research team used a thematic analysis method. Data were coded without necessarily fitting them into a pre-existing coding frame or the researcher's analytic preconceptions.

The 2 research assistants transcribed the data and coded it manually. The researchers then analysed the data manually, examined the transcripts and field notes, and reviewed documents. Using an inductive approach, the researchers identified concepts that emerged and were strongly linked to the data themselves²⁴ and that described the phenomenon under investigation.²⁵ These concepts were further analysed to identify their similarities and differences; subsequently, they were grouped together to form more precise categories that were later organised into themes based on the research objective. Manual analysis was done for documents to clarify the themes and hence understand the emerging themes in the context of Tanzania.

Ethical Consideration

Ethical clearance to conduct this study was obtained from the Medical Research Coordinating Committee through the National Institute of Medical Research. After obtaining ethical approval, permission to carry out the study was obtained from the regional and district officials. The district officials introduced the research teams to the wards and villages. Written informed consent was obtained from all the respondents. The consent form contained information on the objective of the study, procedures of the study, rights to withdraw, confidentiality, and feedback of results. The

instruments were translated into the local language, Kiswahili, and interviews and FGDs were held in private.

RESULTS

Barriers at the District Level

A total of 18 interviews were conducted with service providers, namely the district hospital secretary, in-charges of health facilities, heads of care and treatment clinics, laboratories, and reproductive and child health units from the selected health facilities.

The research team conducted a thematic analysis of the data. Seven themes emerged showing institutional barriers hindering the provision of PITC services at health facilities: a shortage of tools and supplies for providing testing and counselling services, a shortage of health workers at the facility level, overcrowding of clients, inadequate number of trained service providers on PITC, low motivation amongst workers, inadequate space for providing PITC, and harsh language used by some service providers.

Shortage of Tools and Supplies for Providing Testing and Counselling Services

Our findings showed that the district health system lacks adequate tools and supplies for HIV testing. Key informants reported a shortage of diagnostic kits and buffers (reagents) as a common barrier at most of the health facilities visited in the 2 districts. In some places, even consent forms were in low supply. Key informants reported that this shortage of tools and supplies discourages clients from coming to facilities where they do not receive adequate services.

Sometimes we have many clients until we run out of diagnostic kits or reagents . . . we may stay for a month or more without kits or reagents . . . —Key informant, district hospital

Another respondent said:

The government should first address the challenge of insufficient diagnostic kits and reagents if it is interested to ensure HIV test is done to all eligible clients including children . . . for us we play our part if there are adequate resources. —Key informant, district hospital

Shortage of Health Workers at the Facility Level

Findings from most of the key informants show that there was a shortage of staff to deliver the PITC services in many health facilities in the 2 study districts.

We have a severe shortage of staff here . . . and this hospital although is located at a town council we are doing a job like a regional hospital . . . some of our clients come from areas that are outside our catchment area . . . —Key informant, health centre

Another study informant expressed the following:

Sometimes we are asked to postpone our annual leave because of shortage of workforce. . . . This is a major challenge. —Key informant, health centre

A midterm evaluation of the 2008–2013 Human Resources for Health Strategic Plan in 2013 showed that the country had only 44% of the health workers required to serve the total population of the country.²² This lack of availability was asymmetrically distributed between rural and urban areas; although rural areas contained more than 75% of the total population, they had only 55% of the health workers needed to serve the population.²³

Overcrowding of Clients

Key informants reported overcrowding of clients in some health facilities, which is caused mainly by the shortage of staff. The problem is worse in some health facilities, especially those based in town centres where other clients come from nearby districts and even regions.

Sometimes we are facing a problem of overcrowding of clients to the extent that a service provider cannot remember to initiate testing and counselling service for the infants or even children under 5 . . . because we do not even get time for lunch break . . . —Key informant, district hospital

One respondent also stated that the problem of overcrowding of clients also exist in health facilities situated in rural areas.

You know . . . because of shortage of staff in this facility, in most cases you find us overwhelmed with clients making it difficult to provide all services . . . —Key informant, health centre

Inadequate Number of Trained Service Providers on PITC

Key informants in all districts were concerned with the problem of having service providers who are not trained on PITC or exposed to counselling services. This situation was described as a barrier toward effective provision of PITC services in some of the health facilities as expressed below by key informants:

. . . it is not easy for the facility to provide effective provider-initiated testing and counselling services when there is not enough staff trained for this service or oriented for such services . . . —Key informant, health centre

. . . job training has never been enough. Staff must attend at least short courses to gain more knowledge and skills to initiate counselling and HIV testing services for their clients . . . —Key informant, district hospital

Low Motivation Amongst Workers

Findings from this study revealed that the majority of health workers in departments where PITC is delivered had low motivation due to a number of factors. Some of the factors

included work overload, lack of regular refresher courses, inadequate supplies for rendering services, and low remuneration. One of the key informants had this to say:

We are only 2 in this department, today my colleague is sick. I have so many clients as you saw . . . I have been here for more than 5 years but I have not attended even a 1-week seminar . . . I just work because I have no alternative work, but I am really tired . . . —Key informant, district hospital

Health workers in public health facilities are experiencing a heavy workload, which contributes to low motivation, as expressed by a study participant:

Work overload is greatly demoralising me. The government should recruit more staff if it wants to improve the provision of services . . . —Key informant, district hospital

Inadequate Space for Providing PITC

It was reported that since PITC is provided in all departments, a shortage of rooms in many health facilities in all districts reduces the level of privacy to the clients. In 1 of the districts, key informants said that some of the doctors' consultation rooms accommodate 2 or more clinicians who provide services at the same time. In this regard, a key informant expressed the following:

. . . availability of rooms is a barrier; we have very few buildings here and some of the rooms accommodate more than 1 doctor . . . now in such scenario privacy is always compromised, this may limit others from up-taking PITC services . . . —Key informant, district hospital

As you can see here, we have very limited space here. How can you ensure privacy when interacting with clients . . .? —Key informant, health centre

Harsh Language Used by Some Service Providers

Another barrier for the effective provision of PITC services is that some service providers use harsh language when interacting with clients. FGD participants described harsh language as a demotivating factor for clients to accept PITC services in some of the facilities.

Some service providers treat clients with negative attitude and they use harsh language to the extent that some patients hesitate to undertake the requested HIV testing . . . —Recently delivered mother, FGD

There are few service providers who are not user friendly . . . but we have no alternative health facility . . . no option. —Recently delivered mother, FGD

Barriers at the Community Level

Study participants were asked to give their views about community barriers hindering the use of PITC in their areas.

The analysis of the findings generated 3 main themes: low understanding of community members about the importance of HIV testing for children, reluctance of male partners to accompany their partners to the health facility, and stigma associated with HIV/AIDS.

Low Understanding of Community Members About the Importance of HIV Testing for Children

Study participants mentioned that some community members, both men and women, have low understanding about the importance of HIV testing for infants and children under 5. The FGD participants reported a lack of health promotion campaigns about the need for HIV testing for children.

... majority of us including our partners are not aware of the importance of HIV testing for our infants. Maybe there should be more health campaigns to sensitise the community members about HIV testing to our children. —Recently delivered mother, FGD

Reluctance of Male Partners to Accompany Their Partners to the Health Facility

Key informants reported that the provision of PITC could be more effective if male partners could accompany their female partners when visiting health facilities. They noted that men were reluctant to attend reproductive and child health clinics, making it difficult to initiate PITC services to women only. Low male involvement makes counselling difficult when disclosing results to the partner, as reported by a key informant:

... most of the time you have the woman herself visiting a health facility, the man is not there ... it is very hard to counsel the woman to disclose her status or child status to her partner ... It could be easy if they were together ... —Key informant, health centre

Recently delivered mothers in FGDs reported similar findings that low male involvement in reproductive and child health services hinders women's efforts in accessing PITC services. This concern was well narrated by 2 recently delivered mothers:

... most men here do not want to accompany you to the clinic ... they tell you when you are tested it is the same as I have also been tested ... it therefore becomes difficult to tell him when you are tested HIV positive, as sometimes he will beat you claiming that you are the source ... —Recently delivered mother, FGD

If our husbands could escort us to the clinic, they could participate in the counselling session and HIV testing and accepting the results could be easy to both of us ... —Recently delivered mother, FGD

Stigma Associated With HIV/AIDS

Recently delivered mothers who participated in the FGDs said the main barrier for them to accept PITC is stigma

associated with HIV/AIDS in the community. Most of the women are afraid of how fellow community members would look at them when they know they are HIV positive. They reported further that they fear being segregated, a situation that would affect them psychologically. Recently delivered mothers who participated in the FGDs expressed that stigma still persists in the community because some of the community members have low knowledge about HIV/AIDS.

Stigmatising HIV-positive people is a result of low level of knowledge about HIV/AIDS, provision of HIV education should be an ongoing intervention ... —Recently delivered mother, FGD

Another recently delivered mother said that stigma associated with HIV/AIDS is accelerated by unethical health workers who sometimes disclose a patient's HIV-positive diagnosis to community members.

Other service providers announce that someone is HIV positive. This is why some people are hesitant to easily accept HIV test ... —Recently delivered mother, FGD

DISCUSSION

This study aimed to identify health system barriers at the district and community levels that hinder the provision of PITC for infants and children under 5. At the district level, the study found a number of barriers hindering the provision of PITC services, most of which were found at the health facility level. Shortages of adequate service providers, lack of PITC training to the available service providers, and lack of adequate diagnostic kits were amongst the leading barriers. On one hand, shortages of human resources, trained personnel, and diagnostic kits for PITC are contrary to the Tanzania National HIV/AIDS Policy, which emphasises the availability and accessibility of HIV testing and counselling services to all people in the country.²⁶ On the other hand, the national guidelines for HIV counselling and testing in Tanzania stipulate clearly that PITC could only be effective if there are adequate service providers who are trained in PITC.¹⁵

A study on knowledge, attitudes, and acceptability to PITC from the patients' perspectives in Moshi and Rombo districts in Tanzania revealed similar findings – that lack of adequate human resources in the health facilities hinder the provision of PITC services.²⁷ A similar study conducted in Mbeya Region in Tanzania examined perceived barriers and attitudes of health care providers toward PITC and revealed that lack of trained staff may affect the provision of PITC services.²⁸ In Zimbabwe, a study on barriers to PITC for children in a high HIV-prevalence setting reported that health workers lacked training on counselling for children and guardians, which affected the provision of PITC services. This study emphasised that service providers should be trained on PITC, paying particular attention to counselling male guardians, who are less likely to engage with health

care services.²⁹ In Flanders, Belgium, Manirankunda and colleagues reported that lack of expertise in sexuality counselling is a challenge for PITC services to sub-Saharan African migrants.³⁰

In this study, we found that lack of private space and shortages of PITC guidelines for paediatrics, rapid test instruction manuals, and rapid test kits hindered the effective implementation of PITC services. A study in Malawi reported similar findings showing that lack of privacy in the clinics limited uptake of PITC services for infants.³¹ Lack of adequate resources required to implement PITC has also demoralised service providers, thus constraining effective provision of PITC services. A number of studies^{27,32,33} have discussed the importance of incentives for health workers, such as good salaries and training, to increase motivation and commitment and ultimately improve performance. This implies that lack of incentives amongst health workers leads to poor work performance. Lack of HIV testing kits or shortages of kits in various hospital laboratories has been reported elsewhere including Zimbabwe.²⁹

Olmen and colleagues suggest that for the health system to contribute significantly toward improving the performance of health service delivery, there must be an adequate and competent health workforce, good governance, and sufficient resources.³⁴ Furthermore, according to WHO, the government, through health sectors, needs to put in place health financing mechanisms that can ensure access to health services for all people, which in turn help to protect community members from disastrous health expenditure.³⁵

At the community level, understanding and acceptance of parents and caregivers to undertake HIV testing for their children were mentioned as important factors that may influence the provision of PITC services in the study area. Similar findings were also reported from Zimbabwe, showing that some caregivers declined or did not give consent when service providers asked them to undertake HIV test for the children.²⁹

Furthermore, this study shows the surprising finding that stigma associated with HIV/AIDS persists in the community, despite that it has been more than 3 decades since the implementation of interventions focused on reducing stigma. In this study it was reported that HIV/AIDS-related stigma hinders the provision of PITC services. These findings are in agreement with other studies,^{27,36} which underscores that HIV stigma is a major barrier to implementation of HIV interventions in communities, including PITC services.

Moreover, this study found that low male involvement in attending reproductive, maternal, and child health services has limited the provision of PITC services in various health facilities. Because men are still dominant in making decisions pertaining to various household or family matters in most communities in Tanzania, their low involvement in reproductive and child health has a substantial effect on the uptake of PITC services in the community. A number of studies³⁷⁻³⁹ have pointed out a similar barrier indicating that use of HIV testing and counselling services in most health

facilities is low because men do not accompany their female partners to the health facilities.

CONCLUSION

This study concludes that the provision of PITC services at the district level faces a number of barriers, including lack of trained service providers, inadequate number of health workers, lack of adequate space, and shortages of HIV testing kits. At the community level, the provision of PITC services is constrained by lack of understanding and acceptance amongst parents and caregivers to undertake HIV testing for their children, low male involvement in seeking PITC services, and persistence of stigma associated with HIV/AIDS in the community. The study underscores that for the health system to achieve the desired goals of improving PITC services, the government needs to strengthen district- and community-level health systems through recruiting and deploying adequate skilled health workers, training service providers on PITC, mobilising adequate financial resources, and supplying sufficient medicine and other medical supplies. The government should also play an active stewardship role to ensure that the district health system implements interventions to increase awareness and understanding of the importance of PITC services and to reduce or eliminate stigma associated with HIV/AIDS in the community.

Acknowledgements: The authors gratefully acknowledge National Institute for Medical Research through its research grants – Global Fund Round 8-HIV Initiatives – for supporting this study. The authors also acknowledge Tanzania Action for Coastal Education Advocacy, which managed the funding and administration of the project. We are also grateful to the Njombe regional and Makete rural and Njombe urban district, ward, and village administrative and health offices for their administrative support during fieldwork preparation and data collection, the research assistants for the data collection, and the key informants from the district, facility, and community levels who provided information for this study.

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Peer Reviewed

Competing Interests: None declared.

Received: 29 Oct 2016; **Accepted:** 13 Jul 2017

Cite this article as: Frumence G, Nathanaeli S. Health System Barriers to Provider-Initiated HIV Testing and Counselling Services for Infants and Children: A Qualitative Study From 2 Districts in Njombe, Tanzania. *East African Health Res J*. 2017;1(2):123–129. <https://doi.org/10.24248/EAHRJ-D-16-00359>

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Sociodemographic and Other Characteristics Associated With Behavioural Risk Factors of HIV Infection Among Male Mountain-Climbing Porters in Kilimanjaro Region, Tanzania

Jenipher E Lyamuya,¹ Bernard Njau,² Damian J Damian,² Tara B Mtuy^{2,3}

¹African Medical and Research Foundation, Moshi, Tanzania; ²Kilimanjaro Christian Medical University College, Moshi, Tanzania; ³London School of Hygiene & Tropical Medicine, London, UK
Correspondence to Tara Mtuy (tara.mtuy@lshtm.ac.uk).

ABSTRACT

Background: Alcohol consumption, marijuana use, unprotected sex, and multiple concurrent sexual partnerships are prevalent among youth globally. These factors are regarded as important behavioural risk factors for HIV infection. The aim of this study was to assess the sociodemographic and other characteristics associated with behavioural risk factors of HIV infection among male mountain-climbing porters working on Mount Kilimanjaro in Tanzania.

Methods: This cross-sectional study enrolled a representative sample of 384 male mountain-climbing porters from 7 tour companies in the Kilimanjaro region using a multi-stage sampling technique. Local interviewers completed a structured questionnaire with porters in the local language, Kiswahili. The questionnaire covered demographics, alcohol and marijuana use, sexual history, sexual partners, and condom use. In-person interviews were completed between April and May 2013. Univariate and bivariate analysis were used to describe data and determine significant predictors of behavioural risk factors of HIV infection.

Results: Of 384 participants, 381 (99.2%) were sexually experienced, 353 (92.6%) were sexually active, and 168 (44.1%), reported condom use at last sex. The prevalence of ever-use of alcohol was 62%, and 68% of participants reported being current alcohol users. The prevalence of ever-use of marijuana was 15%, and 49% of participants reported being current marijuana users, with 12% reporting daily use. Age, marital status, working duration as a porter, transactional sex practices, and number of concurrent sexual partners were factors that were significantly associated with unprotected sex, alcohol consumption, and marijuana use ($P < .05$).

Conclusions: Age, marital status, working duration, transactional sex practices, and number of concurrent sexual partners were significantly associated with unprotected sex, alcohol consumption, and marijuana use, among porters in this setting. The findings suggest the need for efforts to motivate sexually active male porters to engage in HIV prevention interventions, including condom use and reduction of multiple concurrent sexual partners, transactional sexual practices, alcohol consumption, and marijuana use.

INTRODUCTION

HIV and AIDS remain a worldwide global health problem with an estimated 36 million people living with HIV globally.¹ Sub-Saharan Africa is the region most affected by HIV/AIDS, contributing 75% of the global HIV burden.¹ Preventing and controlling HIV/AIDS among youth aged 15–24 years is a current global challenge. According to the United Nations Population Fund (UNFPA), the youth population has surpassed 1 billion globally—the largest in human

history.² In Tanzania, almost two-thirds (65%) of the population is under the age of 24 years, and youth are affected most by HIV/AIDS with an overall prevalence of 7.9%.^{3,4}

One of the major drivers of the high HIV prevalence among youth is early sexual activity. For example, a substantial proportion (85%) of unmarried youth in Tanzania are sexually experienced with low (12.2%) condom-use practices.⁵ Alcohol consumption and marijuana use are also becoming a major global public health problem, particularly among youth. According

to the World Health Organization (WHO), alcohol consumption and marijuana use contribute 4% of the disability-adjusted life years of the global burden of disease.⁶⁻⁹ In sub-Saharan Africa, approximately 25% of young people are current drinkers and reported to consume 35 litres of pure alcohol annually.^{10,11} In Tanzania, the proportion of substance use, including alcohol consumption, is increasing, with 17.2% of urbanite youth current users.¹⁰ The high alcohol consumption among youth is compounded with risky sexual behaviours, such as multiple concurrent sexual partnerships and low or inconsistent condom use.¹⁰

In Kilimanjaro region, public health officers have identified 2 high-risk populations for HIV – female bar workers and male mountain-climbing porters. While several studies in this area have documented HIV risk behaviours among female bar workers^{12,13} and among youth in different settings,^{5,16} there is limited information pertaining to risk characteristics specifically of young male mountain-climbing porters, who may share many risk characteristics with other high-risk groups, such as long-distance truck drivers,^{17,18} fishermen,¹⁹⁻²¹ miners,^{22,23} and migrant farm workers.²⁴

The estimated 17,000 porters of Mount Kilimanjaro are between the ages of 18 and 45 years, but are predominantly young men who are very mobile and face volatile income cycles.¹⁴ Evidence suggests that people who work in difficult environments, have seasonal jobs, and earn a higher-than-average living wage are at high risk of overspending money on alcohol and marijuana and are more likely to have multiple sexual partners. Such risky behaviours may lead to unsafe sex practices, posing a risk of HIV infection.¹⁵

The aim of this study was to assess the sociodemographic and other characteristics associated with behavioural risk factors of HIV infection among male mountain-climbing porters in Kilimanjaro region, Tanzania. The findings from this study will contribute to knowledge of risk characteristics of male porters of Mt. Kilimanjaro and may assist knowledge brokers, policy makers, and HIV prevention interventionists to develop effective strategies to influence behaviour change for prevention of HIV transmission among this high-risk group in this study setting.

METHODS

Study Population and Protocol

We conducted a cross-sectional study designed to assess sociodemographic and other characteristics associated with behavioural risk factors for HIV infection among male mountain-climbing porters in the Kilimanjaro region. According to a conversation with a representative from the Kilimanjaro Porters Assistance Project (February 2013), it is estimated that a total of 17,000 porters are registered with the 3 Kilimanjaro porters unions. A multistage sampling technique was used to obtain the estimated sample of participants. In the first stage, we randomly selected 7 of 14 tour

companies. In the second stage, we obtained a list of all porters working in the 7 tour companies from the tour companies' management and then used sampling proportionate to size technique to select a random sample of 384 participants. Sample size calculation was based on an HIV prevalence of 50%, as no other study has been done on behavioural risk factors for HIV among porters. Local interviewers were trained on obtaining informed consent, maintaining confidentiality, following interview procedures, and completing the questionnaire. Study participants, recruited between April and May 2013, completed in-person interviews in Kiswahili with the interviewers.

Measures

Alcohol use was assessed by 11 questions whereby 4 questions were adapted from CAGE questionnaire.²⁵ Marijuana use was assessed by 6 structured questions and a composite score was calculated. Participants self-reported sociodemographic information included age, marital status, highest level of completed education, and income. Condom use was assessed by asking participants if they had had sex without a condom during the last sexual intercourse (1=yes; 2=no). A response of 'yes' indicated sexual risk behaviour. Transactional sex practices were assessed by asking participants if they had exchanged/paid/received a gift for sex during the last 6 months (1=no; 2=yes, received; 3=yes, given; 4=yes, received and given). Number of sexual partners was assessed by asking participants the number of casual sexual partners they had during the last 3 months. The number of any sexual partners was dichotomised and coded: 'no multiple sexual partners' (≤ 1 sexual partner) or 'multiple sexual partners' (> 1 sexual partner).

Statistical Analyses

Data were entered, cleaned, and analysed using Statistical Package for Social Science (SPSS) version 18 (SPSS, Inc., Chicago IL). Categorical data were summarised using frequency and percentages, while numerical data were summarised using measures of central tendencies with their respective measures of dispersions. Chi-square test was used to determine statistical associations between categorical variables. Bivariate logistic regression analysis was used to determine sociodemographic and other characteristics associated with behavioural risk factors of HIV infection among study participants. A *P*-value was considered statistically significant at the .05 level.

Ethical Approval

The study and all study activities were approved by the Kilimanjaro Christian Medical University College Research Ethical Review Committee. Written informed consent was obtained. Participants were informed of their voluntariness to participate and freedom to withdraw at any point from the study.

RESULTS

Background Characteristics

Sociodemographic Characteristics

In total, 384 porters were enrolled in the study. The mean age of study participants was 31 years (standard deviation [SD], 6.6) (Table 1). More than half (n=199, 51.8%) of the study participants were married. Most (n=247, 64.3%) had primary education, and 60.2% earned an average monthly income of less than 400,000 Tanzanian Shillings (TZS) during high season (in 2012, US\$1=1,600 TZS). More than half (n=219, 57.0%) of participants had been working as a porter for 3 to 5 years.

Sexual Characteristics

Nearly all participants (n=381, 99.2%) reported being sexually experienced (Table 2). Of the 381 respondents who reported being sexually experienced, 177 (46.5%) reported having sex 1 to 6 days prior to the interview and 353 (92.6%) said they had had sex during the last 3 months. More than half (54.6%) of sexually experienced respondents had had sex with their wives. The majority (85.8%) of participants did not report having sex in exchange for money or a gift during the last 6 months prior to the interview. Out of the 353 sexually active participants who had had sex during the last 3 months, 61 (17.3%) reported multiple concurrent sexual partners. Of the 381 sexually experienced participants, 168 (44.1%) reported condom use during their last sexual intercourse.

Prevalence of Alcohol and Marijuana Use

Of the total 384 participants, 237 (61.7%) reported ever drinking alcohol (Table 3). Of these, 161 (67.9%) reported being current drinkers. Of the 161 current drinkers, 81 (50.3%) reported drinking 2 to 7 times per week and drinking on average 4 bottles (SD, 3; range, 1–25) during a typical day. The majority (n=136, 84.5%) of current drinkers reported drinking beer, and 6 (3.7%) took 6 or more drinks at 1 occasion daily.

Of the 384 participants, 57 (14.8%) reported to have ever used marijuana (Table 4). Mean age at first time using marijuana was 18 years (SD, 5.4). The youngest age reported to have started using marijuana was 7 years while the oldest age was 45 years. The prevalence of reported current marijuana use was 49.1% (n=28). Out of 28 respondents who reported current marijuana use, 12 (42.9%) reported daily use.

Sociodemographic and Other Characteristics Associated With Behavioural Risk Factors of HIV Infection Among Study Participants

The 4 behavioural risk factors of HIV infection in this study were condom use during the last sexual intercourse, having multiple concurrent sexual partners, and use of alcohol and marijuana. Age was significantly associated with all 4 of

TABLE 1. Sociodemographic Characteristics of Respondents (N=384)

Variable	n (%)
Age (years)	
20–29	162 (42.2)
30–39	172 (44.8)
40–49	50 (13.0)
Level of education	
None	2 (0.5)
Primary	247 (64.3)
Secondary	126 (32.8)
Post-secondary	9 (2.3)
Marital status	
Married	199 (51.8)
Single	127 (33.1)
Cohabiting	54 (14.1)
Divorced	3 (0.8)
Widowed	1 (0.3)
Monthly income during high season (TZS) (n=383)	
<200,000	86 (22.4)
200,000–399,000	145 (37.8)
400,000–599,000	69 (18.0)
600,000–799,000	48 (12.5)
800,000+	36 (9.4)
Duration working as a porter (years)	
≤2	32 (8.3)
3–5	219 (57.0)
6–8	92 (24.0)
9–11	35 (9.1)
12 +	6 (1.6)
Duration of work, years, mean (standard deviation)	5.9 (4.6)

these behavioural risk factors ($P<.05$): older participants were more likely than younger participants (20–29) to have not used condoms at last sex (30–39 year age group: odds ratio [OR] 4.60; 95% confidence interval [CI],

TABLE 2. Sexual Characteristics of Study Participants (N=384)

Variable (n)	n (%)
Ever had sex	
Yes	381 (99.2)
No	3 (0.8)
Last time had sex (381)	
1–6 days ago	177 (46.5)
7 days ago	65 (17.1)
1–11 months ago	101 (26.5)
≥12 months ago	38 (10.0)
Relationship to the woman with whom had last sex (381)	
Wife	208 (54.6)
Fiancée	35 (9.2)
Regular partner	77 (20.1)
Causal partner	54 (14.2)
Tourist	2 (0.5)
Commercial sex worker	5 (1.3)
Place you first met this woman (381)	
Own/friend’s house	55 (14.4)
Family event	9 (2.4)
Bar/hotel	22 (5.8)
Park/company	11 (2.9)
Church	18 (4.7)
Other places	266 (69.8)

Continued

TABLE 2. Continued

Variable (n)	n (%)
In last 6 months, exchanged/paid money/gift for sex (381)	
No	327 (85.8)
Yes, received	7 (1.8)
Yes, given	39 (10.2)
Yes, received and given	8 (2.1)
Relationship to the woman with whom paid/gave/received money/gift for sex (330)	
Tourist	5 (9.3)
Commercial sex worker	12 (22.2)
Bar/hotel worker	13 (24.1)
Sex partner/friend	24 (44.4)
Had multiple sexual partners in the past 3 months (353)	
Yes	61 (17.3)
No	292 (82.7)
Used condom at last sex (381)	
Yes	168 (44.1)
No	215 (55.9)

2.76–7.73; 40–49 year age group: OR 20.20; 95% CI, 4.75–86.10) but less likely to have multiple concurrent sexual partners (OR 0.39; 95% CI, 0.21–0.73 vs. OR 0.61; 95% CI, 4.75–86.10, respectively). Similarly, older participants were more likely than younger participants to consume alcohol (OR 1.71; 95% CI, 1.12–2.72 vs. OR 1.63; 95% CI, 0.84–3.16, respectively) and to use marijuana (OR 0.54; 95% CI, 0.30–0.99 vs. OR 0.25; 95% CI, 0.07–0.85, respectively).

Marital status was significantly associated with condom use during last sexual intercourse, having multiple concurrent sexual partners, and marijuana use ($P < .05$). Compared

with married participants, those who were single were less likely to have had unprotected sex during last sexual intercourse (OR 0.06; 95% CI, 0.03–0.11), but were more likely to have multiple concurrent sexual partners (OR 3.27; 95% CI, 1.76–6.07) and to use marijuana (OR 5.76; 95% CI, 2.78–11.92).

In general, the longer respondents worked as a porter, the higher the likelihood of them not using condoms as last sex ($P < .001$).

Receiving money or gifts for sex during the last 6 months was significantly associated with having unprotected sex during the last sexual intercourse (OR 1.49; 95% CI, 1.11–2.25). In addition, receiving or giving money or gifts for sex was significantly associated with having multiple concurrent sexual partners and with marijuana use ($P < .05$).

Furthermore, the number of any sexual partners in the past 3 months was significantly associated with unprotected sex during the last sexual intercourse ($P = .001$). Respondents with more than 1 sexual partner were more likely not to

TABLE 3. Prevalence of Alcohol Use Among Study Participants (N=384)

Variable (n)	n (%)
Ever drank alcohol (384)	
Yes	237 (61.7)
No	147 (38.3)
Currently drinking alcohol (237)	
Yes	161 (67.9)
No	76 (32.1)
Frequency of drinking (161)	
Monthly or less	15 (9.3)
2-4 times a month	65 (40.4)
2-3 times a week	57 (35.4)
4+ times a week	24 (14.9)
Type of alcohol (161)	
Beer	136 (84.5)
Liquor	3 (1.9)
Mbege (local brew)	21 (13.0)
Both	1 (0.6)
Frequency of having 6 or more drinks on 1 occasion (161)	
Never	106 (65.8)
Once per month	25 (15.5)
2-3 times per month	15 (9.3)
Weekly	9 (5.9)
Daily/almost daily	6 (3.7)
Number of drinks taken on a typical day, mean (standard deviation) (161)	4.12 (3.14)

TABLE 4. Prevalence of Marijuana Use Among Study Participants (N=384)

Variable (n)	n (%)
Ever used marijuana (384)	
Yes	57 (14.8)
No	327 (85.2)
Currently using marijuana (57)	
Yes	28 (49.1)
No	29 (50.9)
Age first started using marijuana (57)	
≤10	3 (5.3)
11-20	42 (73.7)
21-30	11 (19.3)
31-40	0 (0.0)
41+	1 (1.8)
Usual frequency of using marijuana (28)	
Every day	12 (42.9)
2-3 times a week	9 (32.1)
2-3 times a month	5 (17.9)
Once in a month	2 (7.1)

use condoms in their last sexual intercourse (OR 1.38; 95% CI, 0.21-0.67).

Level of income was not significantly associated with any of the 4 studied behavioural risk factors ($P > .05$).

DISCUSSION

The study findings have shown that certain sociodemographic characteristics, as well as paying or receiving money or gifts for sex, are associated with behavioural risk factors for HIV infection among porters in Kilimanjaro region, Tanzania. Overall, the majority of porters were sexually

experienced, more than half (56%) reported they did not use a condom during their last sexual intercourse, and almost one-fifth (17%) reported having multiple concurrent sexual partners.

Overall, older respondents were more likely to have not used condoms, so unprotected sex increased with increasing age, but the likelihood of having multiple concurrent sexual partnerships decreased with increasing age. The most probable explanation to these observations may be due to the fact that young male porters tend to use condoms because they also engage in multiple concurrent sexual partnerships, hence perceive themselves being at high risk of HIV infection.^{5,26,27} In contrast, older male porters are less likely than older porters to use condoms because they may be in a regular sexual partnership. Additionally, this observation of high condom use among younger participants can possibly be due to exposure to sensitisation messages on condom use.⁴ This finding is in contrast with findings from previous investigators on youth's sexual activity in Tanzania, who found that condom use was higher among older respondents.^{4,16}

TABLE 5. Sociodemographic Characteristics Associated With Behavioural Risk Factors for HIV Among Study Participants (N=384)

Characteristics	Total (n)	Behavioural Risk Factors			
		Did Not Use Condom During Last Sex OR (95% CI)	Had Multiple Concurrent Sexual Partner OR (95% CI)	Use Alcohol OR (95% CI)	Use Marijuana OR (95% CI)
Age (years)					
20–29	162	1	1	1	1
30–39	172	4.60 (2.76, 7.73)	0.39 (0.21, 0.73)	1.71 (1.12, 2.72)	0.54 (0.30, 0.99)
40–49	50	20.20 (4.75, 86.10)	0.61 (4.75, 86.10)	1.63 (0.84, 3.16)	0.25 (0.07, 0.85)
Marital status					
Married	199	1	1	1	1
Single	127	0.06 (0.03, 0.11)	3.27 (1.76, 6.07)	1.01 (0.64, 1.50)	5.76 (2.78, 11.92)
Cohabiting	54	0.47 (0.20, 1.12)	1.67 (0.72, 3.89)	1.32 (0.70, 2.48)	5.42 (2.27, 12.95)
Divorced/widowed	4	0.19 (0.02, 2.17)	4.00 (0.35, 45.94)	NC	8.55 (0.72, 10.66)
Monthly income during high season (TZS)					
<200,000	86	1	1	1	1
200,000–399,000	145	1.60 (0.65, 3.90)	0.68 (0.22, 2.12)	1.62 (0.69, 3.78)	1.55 (0.42, 5.71)
400,000–599,000	69	1.36 (0.58, 3.16)	0.79 (0.27, 2.29)	0.95 (0.86, 4.40)	1.60 (0.45, 5.67)
600,000–799,000	48	2.52 (0.81, 7.84)	1.04 (0.29, 3.75)	1.99 (0.72, 5.47)	0.71 (0.13, 3.80)
800,000+	36	3.43 (1.07, 11.01)	1.16 (0.34, 3.97)	1.68 (0.63, 4.49)	1.11 (0.24, 5.09)
Duration working as a porter (years)					
≤2	32	1	1	1	1
3–5	219	2.61 (1.48, 4.60)	1.19 (0.54, 2.59)	1.83 (1.07, 3.14)	0.54 (0.26, 1.15)
6–8	92	3.68 (1.76, 7.70)	1.31 (0.54, 3.18)	4.14 (2.03, 8.44)	0.92 (0.40, 2.10)
9–11	35	4.24 (1.77, 10.13)	1.71 (0.67, 4.36)	1.36 (0.67, 2.76)	0.63 (0.23, 1.72)
12+	6	4.03 (1.65, 9.83)	0.33 (0.07, 1.58)	4.03 (1.65, 9.83)	
In last 6 months, exchanged/paid money/gift for sex					
No	327	1	1	1	1
Yes, received	7	1.49 (1.11, 2.25)	3.35 (1.62, 17.97)	0.49 (0.11, 2.25)	2.79 (1.52, 14.85)
Yes, given	39	1.91 (0.90, 4.06)	11.52 (5.49, 24.16)	1.91 (0.90, 4.06)	3.10 (1.46, 6.59)
Yes, received and given	8	1.10 (0.26, 4.68)	13.96 (3.19, 61.17)	1.10 (0.26, 4.68)	0.99 (0.12, 8.31)
Number of any sexual partner in the past 3 months					
≤ 1	292	1	-	1	1
More than one	61	1.38 (1.21, 0.67)	-	1.51 (0.83, 2.74)	1.99 (1.00, 3.96)
Total		73.0% (n=278)	17.3% (n=61)	61.7% (n=237)	14.8% (n=57)

Abbreviations: CI, confidence interval; NC, not calculated; OR, odds ratio.

Marital status was associated with unprotected sex, multiple concurrent sexual partnerships, and marijuana use. The likelihood of condom use was higher among unmarried participants than their married counterparts. Furthermore, unmarried participants were more likely to have multiple concurrent sexual partners and to smoke marijuana. This finding is consistent with behavioural studies assessing motivating factors associated with condom use among youth, which found that respondents who perceived higher susceptibility to HIV infection through their risky behaviours were more likely to report condom use.^{5,16}

Transactional sex was associated with unprotected sex, having multiple concurrent partners, and marijuana use. Porters who reported to have paid and/or received money or a gift in exchange for sex were more likely to report unprotected sex and to smoke marijuana. The finding that transactional sexual practice was associated with unprotected sex is consistent with several studies among youth in different settings. Existing evidence on HIV infection in sub-Saharan Africa shows that transactional and transgender sexual practices are 2 major drivers for the high HIV prevalence in the general population, including youth.¹ Additionally, the observed risk behaviour of mixing unprotected sexual practices and marijuana smoking underscores the importance of skills development for safer sex among porters in this setting. An intervention designed to increase motivation for condom use while addressing the mixed-risk behaviours may be an effective approach.^{1,28,29}

Finally, the number of concurrent sexual partners in the last 3 months was associated with non-condom use among porters. Porters who reported more than 1 concurrent sexual partner during their last sexual intercourse were more likely to engage in unprotected sex. This observation is consistent with studies on condom use among youth,^{1,30,31} and raises concern that porters in this setting are at high risk of contracting HIV. Indeed, low rates of condom use coupled with high rates of multiple concurrent sexual partners may increase the spread of HIV among male porters.

Limitations

Although this study addressed a number of behavioural risk factors associated with HIV infection among Kilimanjaro mountain porters, some risk factors were not explored. This included knowledge of HIV transmission and prevention, sexual practices, HIV status, and history of other sexually transmitted diseases. Further studies addressing these risk factors are important to better understand this at-risk population for HIV.

This study has other certain limitations. First, the cross-sectional study design that we used is not adequate for measuring the directionality of associations found and,

therefore, cannot account for potential confounders. Second, we asked respondents to recall events that had occurred in the distant past. Recall bias may be a limitation, particularly among older porters who may be unable to remember the exact timing of their early sexual activities. Third, the study relied on self-reports. It is known that the self-reported behaviour is subject to reporting bias, which may overestimate or underestimate the effects of association and the validity of the findings. In particular, due to marijuana use being illegal in Tanzania, it was likely under-reported. Finally, given that this study was conducted in a particular location and among male porters, it may not be applicable to other settings or the general population. Nevertheless, the study findings provide an important insight into male mountain climbing porters' behavioural risk factors for HIV infection.

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Peer Reviewed

Competing Interests: None declared.

Received: 23 Jun 2017; **Accepted:** 4 Aug 2017

Cite this article as: Lyamuya JE, Njau B, Damian DJ, Mtuy TB. Sociodemographic and Other Characteristics Associated With Behavioural Risk Factors of HIV Infection Among Male Mountain-Climbing Porters in Kilimanjaro Region, Tanzania. *East African Health Res J*. 2017;1(2):130-137. <https://doi.org/10.24248/EAHRJ-D-17-00259>

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