

Brief Original Article

## Herpes Simplex virus type 2 seroprevalence and risk factors among adolescents and youth with HIV-1 in Northern, Tanzania

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### Abstract

**Introduction:** Herpes Simplex virus type 2 (HSV-2) infection is associated with an increased risk of human immunodeficiency virus type 1 (HIV-1) acquisition and transmission. Individuals co-infected with HIV-1 and HSV-2 may have longer lasting, more frequent and severe outbreaks of herpes symptoms. Previous studies have assessed HSV-2 seroprevalence and associated risk factors in adult populations. However, there is limited data on the HSV-2 seroprevalence among adolescents and youth living with HIV-1. The study aimed to determine the HSV-2 seroprevalence and associated risk factors among adolescents and youth living with HIV-1 at referral hospital setting in Northern Tanzania.

**Methodology:** A cross-sectional survey was conducted between February and July 2017 among HIV-1-infected individuals aged 10-24 years attending the Child-Centred Family Care Clinic at Kilimanjaro Christian Medical Centre. Blood specimens from 180 individuals were collected for ELISA-based detection of HSV-2 antibodies. Associations between risk factors and HSV-2 seroprevalence were analysed by univariate and multivariate logistic regression models.

**Results:** The overall HSV-2 seroprevalence was 18% (32/180). A significant HSV-2 seroprevalence was noted among adolescents and youth, who reported having had sexual intercourse than those who never had sexual intercourse (28.9% vs 13.3%,  $p = 0.02$ ). Youths aged 20-24 had six folds higher risk of HSV-2 seroprevalence compared to those aged 10-14 years (AOR = 5.97 95% CI 1.31 – 27.19,  $p = 0.02$ ).

**Conclusions:** Our study found that HSV-2 seroprevalence increased by age among adolescents and youth living with HIV-1. Age-specific approaches might play an important role in interventions targeting HSV-2 infection.

**Key words:** Herpes Simplex Virus type 2; human immunodeficiency virus; adolescents; youth; Tanzania.

*J Infect Dev Ctries* 2020; 14(4):398-403. doi:10.3855/jidc.11599

(Received 25 April 2019 – Accepted 14 December 2019)

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### Introduction

HSV-2 is a DNA virus of the *Herpesviridae* family, characterized by short replicative cycles, host cell destruction, and the ability to establish lifelong latency in sensory neural ganglia following primary infection [1]. It is mainly transmitted through genital secretions but can also be transmitted from mother-to-child during pregnancy and delivery [2]. HSV-2 is a common cause of genital ulcer infections worldwide [3]. Globally, 417 million people (11%) are estimated to be living with HSV-2 and Sub-Saharan Africa (SSA) accounts for more than 30% of the cases [4,5]. People who are infected with both HIV-1 and HSV-2 may have longer lasting, more frequent and more severe outbreaks of herpes symptoms [6,7]. HSV-2 co-infected with HIV-1 is associated with increased immune activation and

increased plasma and genital HIV-1 viral loads among untreated individuals [4,8,9]. Moreover, HSV-2 causes ulceration or inflammation of the genital tract and thus may increase the infectiousness of HIV-1-infected individuals and the susceptibility of HIV-1-negative persons [6]. It has been shown that HSV-2 infection increases the risk of HIV-1 acquisition and transmission by 2-3 and 4-folds, respectively [10].

A multi-country review has estimated the HSV-2 seroprevalence to be 5- 8% among adolescents and youth worldwide [4]. A study conducted among adolescents and young people aged 12-28 in rural KwaZulu-Natal found seroprevalence of HSV-2 of 2.6% and 10.7% among males and females, respectively [11]. In Western Kenya, Akinyi and colleagues reported HSV-2 seroprevalence of 10.7%

among adolescents living with HIV-1 [12]. Data from these studies indicate that the distribution of the HSV-2 varies with age, geographical location (between regions and countries) and also between population sub-groups [13].

In sub-Saharan Africa (SSA), despite significant gains in reduction of HIV-1 infections among the general population, yet adolescents and youth's HIV-1 incidences remain high due to several factors including engagement in risky sexual behaviour such as early sexual debut (< 15 years of age), unprotected sexual intercourse, multiple sexual partners and other sexually transmitted infections (STI) including HSV-2 [14]. Furthermore, antiretroviral therapy (ART) retention and viral suppression rates have been reported to be relatively lower among adolescents and youth than adults enrolled in ART programmes [15]. For these reasons, adolescents and youth are key populations in planning HIV-1 preventive and control strategies.

In Tanzania, the expansion of ART interventions has increased the likelihood of HIV-1 infected children surviving into adolescence and adulthood [16,17]. Furthermore, a large proportion of new STIs occurs amongst adolescents and young adults who may not be aware that they are infected which can have a negative impact upon their future sexual and reproductive health [18].

HSV-2 studies conducted in Tanzania have focused on adults, mostly women and only a few studies have focused on adolescents and youth [19–22]. Addressing this gap, we envisaged to investigate the seroprevalence and risk factors associated with HSV-2 infection in adolescents and youth infected with HIV-1 attending the Child-Centred Family Care Clinic (CCFCC) at Kilimanjaro Christian Medical Centre (KCMC), Northern Tanzania.

## Methodology

### *Study population*

Study participants were adolescents and youth aged 10-24 years living with HIV-1 attending CCFCC at KCMC hospital. The participants were recruited during their planned attendance at the clinic, where they also have a chance to participate in activities such as sports and reproductive health education. The participants were approached during these social activities.

We collected Sociodemographic and behavioural characteristics using a standardized questionnaire that was supplemented with information retrieved from the participant's files. The information collected included participants age, date of HIV-1 diagnosis, gender, age at first sexual intercourse, frequency of condom usage

in the past 12 months, condom use at last sex encounter, number of sexual partners, history of genital ulcers, education level, type of family, mode of transmission and clinical stage of HIV disease according to the classification by World Health Organization (WHO). The questionnaires were prepared in English and translated to Swahili and then back-translated into English. The questionnaires were pre-tested and revised accordingly before the actual data collection. Following the interviews, questionnaires were translated into English.

### *Laboratory testing for HSV-2*

HSV-2 antibody testing was performed with HSV-2 IgG enzyme-linked immunosorbent assay (ELISA) kit (93% specificity and 100% sensitivity) according to the manufacturer's instructions (Euroimmuno, Medizinische Labordiagnostika AG, Lübeck, Germany). HSV-2 seropositivity was defined as having detectable IgG antibodies to HSV-2. For specimens with inconclusive HSV-2 results, the samples were re-analysed.

### *Statistics*

Statistical analyses were performed using STATA, version 13.0 (Stata Corporation, College Station, Texas, USA). Descriptive statistics were used to summarize the data. Chi-square or Fisher's exact test was used to compare categorical variables and student's t-test was used to compare continuous variables. Univariate and multivariable logistic regression analysis was used to determine the association between HSV-2 seropositivity and potential risk factors. In the Univariate logistic regression, the variables with a p-value of less than 0.2 were included in the Multivariable logistic regression analysis. The results were presented using Odds Ratio (OR) with 95% confidence intervals. All statistical tests were two-tailed and p-values of  $\leq 0.05$  were considered to be statistically significant.

## Results

A total of 180 participants with mean (SD) age 17.5(3.5) years were enrolled in the study. The relatively high proportion of the participants 96 (53.3%) were female. The median age at sexual debut was 18 years (range 9–23 years). According to the patient's files, most of the participants 163 (90.5%) acquired HIV-1 vertically. HSV-2 status varied considerably by demographic and behavioral characteristics of the study population, (Table 1). The overall seroprevalence for adolescents and youth infected with HSV-2 was 18.0% (32/180).

**Table 1.** Socio-demographic and behavioural characteristics of HSV-2 seroprevalence among HIV-1 infected adolescents and youths.

Variable	n (%)	HSV-2 positive, n (%)	P-value	Crude OR (95% CI)	P-value
<b>Gender</b>					
Male	84 (46.7)	13 (15.5)	0.45	Ref-	
Female	96 (53.3)	19 (19.8)		1.35 (0.62-2.93)	0.45
<b>Age group</b>					
10-14 years	50 (27.8)	3 (6.0)	<b>&lt; 0.01</b>	Ref	
15-19 years	91 (50.6)	16 (17.5)		3.34 (0.92-12.09)	0.06
20-24 years	39 (21.6)	13 (33.3)		7.83 (2.04-30.03)	<b>&lt;0.01</b>
<b>Education level</b>					
None	3(1.7)	1 (33.3)	0.89	Ref-	
Primary	73 (40.6)	13 (17.8)		0.43 (0.04-5.14)	0.51
Secondary	89 (49.4)	15 (16.9)		0.41 (0.03- 4.76)	0.47
Tertiary	15 (8.3)	3 (20.0)		0.50 (0.03- 7.54)	0.62
<b>Tribe</b>					
Chaga	98 (54.4)	20 (20.4)	<b>0.16</b>	Ref-	
Pare	24 (13.3)	1 (4.2)		0.16 (0.02-1.33)	0.13
Others	58 (32.2)	11 (19.0)		0.91 (0.39-2.07)	0.83
<b>Ever had sex?</b>					
No	128 (71.1)	17 (13.3)	<b>0.01</b>	Ref-	
Yes	52 (28.9)	15 (28.9)		2.65 (1.20-5.82)	<b>0.02</b>
<b>Ever consumed alcohol*</b>					
Yes	11 (6.1)	28 (16.6)	0.10	Ref-	
No	169 (93.9)	4 (36.4)		2.88 (0.79-10.49)	0.11
<b>History of genital ulcer</b>					
Yes	12 (6.7)	1 (8.3)	0.70	Ref	
No	168 (93.3)	31 (18.5)		2.50 (0.31 -20.15)	0.86
<b>HIV-1 transmission</b>					
Mother to child	163 (90.5)	29 (17.8)	0.65	Ref	
Others	17 (9.4)	3 (17.7)		0.90 (0.27 - 3.67)	0.99
<b>WHO clinical stage</b>					
I	4 (6.6)	1 (7.1)	<b>0.83</b>	Ref-	
II	23 (35.4)	4 (17.4)		0.63 (0.05-7.75)	0.72
III	38 (58.5)	9 (23.7)		0.93 (0.08-10.09)	0.95

HSV-2: Herpes simplex virus type 2, CI: Confidence interval; OR odds ratio. Significant P-values are presented in bold

**Table 2.** Adjusted multivariate analysis of the association between HSV-2 seroprevalence and potential risk factors.

Variable	Response	AOR (95% CI)	P-value
Gender	Male	Ref	
	Female	1.48 (0.64-3.38)	0.36
Ever had sex	No	Ref	
	Yes	1.28 (0.49-3.35)	0.62
Age group	10 -14 years	Ref	
	15 -19 years	2.93 (0.78-11)	0.11
	20-24 years	5.97 (1.31-27.19)	<b>0.02</b>
Alcohol intake	No	Ref	
	Yes	2.31 (0.57-9.34)	0.24
Tribe	Chagga	Ref	
	Pare	0.26 (0.03-2.15)	0.21
	others	1.24 (0.51-3)	0.63

AOR: adjusted odds ratio; ref-: reference CI: confidence interval. Significant P-values are presented in boldface

The HSV-2 seropositivity for adolescent and youth who reported to have had sexual intercourse were (28.9%) and those who had never had sexual intercourse (13.3%,  $p = 0.01$ ). The univariate analysis showed that youth aged 20 - 24 had an eight folds risk of HSV-2 seropositive compared to adolescents aged 10 - 14 years (OR=7.83, 95% CI 2.04-30.03;  $p = 0.003$ ). Furthermore, individuals who reported to have had sexual intercourse had 3-folds risk of HSV-2 seropositivity relative to those who never had had sexual intercourse (OR = 2.65, 95% CI = 1.20 - 5.82;  $p = 0.02$ ), (Table 1). When the multivariate analysis was adjusted for gender, ever had sex, tribe, alcohol intake and age group, youth aged 20-24 years had a significantly higher risk of HSV-2 seropositivity (AOR = 5.97, 95% CI: = 1.31 - 27.19;  $p = 0.02$ ) than other age groups, (Table 2). For the 52 participants who reported to be sexually active, frequency of condom use during sexual act and number of sexual partners did not differ significantly according to HSV-2 serostatus, (Table 3).

### Discussion

HIV-1 transmission in adolescents and youth is still a challenge particularly in developing countries including Tanzania [23]. Sexually transmitted diseases such as HSV-2 is among the significant risk factors for HIV transmission [10]. We found an HSV-2 seroprevalence of 18% in adolescents and youth, relatively higher than that (12.6%) reported in a previous study conducted in Mwanza, Tanzania [24]. The Mwanza study populations were screening of HIV-1 uninfected and it is possible these two study populations might differ also with other factors than HIV-1 infection status contributing to the difference in HSV-2 seroprevalence. A study in the same age group in Western Kenya, reported HSV-2 seroprevalence of 10.7% and 26.7% among adolescents (aged 16-17) and

youth (aged 18-24), respectively [12]. High HSV-2 seroprevalence in HIV-1 infected individuals observed in our study corroborate with existing evidence of a link between the two diseases as reported elsewhere [6,12,25–27].

We also found that adolescents and youth who initiated sexual intercourse early in life had a 55% percent higher risk of HSV-2 than those who initiated sexual intercourse above 15 years old, although the difference was not statistically significant (Table 3). Previous studies have also shown that seroprevalence rates are higher among adolescents and youth females relative to their male counterparts [4,28]. Moreover, HSV-2 seroprevalence was found to increase with increasing age. Hence, targeting adolescents and youth living with HIV-1 for prevention of HIV-1 and HSV-2 could improve prevention and accelerate control of the two infections. Preventing both HSV-2 and HIV-1 infections through different approaches should be prioritized.

It has been previously shown that multiple sexual partners, inconsistent condom use, sexually transmitted infections (STIs), and alcohol consumption are associated with HSV-2 seroprevalence [14,21]. However, in the current study, no significant association was found between condom use, multiple sexual partners and alcohol consumption and HSV-2 seroprevalence. This could be due to the overall relatively small sample size, the limited number of sexually active individuals and difficulties in obtaining reliable data on sexuality as reported elsewhere [29]. Forty percent (12/30) of the HSV-2 seropositive individuals were reported to have inconsistent condom usage during the last 12 months, and 32% (6/19) of the HSV-2 seropositive participants reported to have multiple sexual partners. Both inconsistent condom use and multiple sexual partners are likely to contribute to

**Table 3.** Distribution of HSV-2 among sexually active youth and adolescent living with HIV-1 (n=52).

Variable	n(%)	HSV -2 seropositive, n (%)	P-value	Crude OR (95% CI)	P-value
<b>Condom use last sex</b>			0.27		
Yes	27 (51.9)	6 (22.2)		Ref	
No	25 (48.1)	9 ( 36.0)		0.51 (0.15 - 1.72)	0.28
<b>Frequent of condom use</b>			0.12		
Never	15 (28.8)	2 (13.3)		Ref	
Inconsistency	30 (57.7)	12 (40.0)		4.33 (0.82 - 22.75)	0.08
Consistency	7 (13.5)	1 (14.3)		0.96 (0.08 - 14.41)	0.95
<b>Number of sexual partners</b>			0.74		
One	33 (63.5)	9 (27.3)		Ref	
Two or more	19 (36.5)	6 (31.6)		1.12 (0.38 - 3.73)	0.87
<b>Age at first sexual intercourse</b>			0.51		
Late Initiation >15 years	14 (26.9)	5 (35.7)		Ref	
Early Initiation ≤ 15 years	38 (73.0)	10 (26.3)		1.55 (0.42 - 5.77)	0.51

HSV-2: Herpes simplex virus type 2.

both HSV-2 and HIV-1 transmission. None of the other factors investigated (including participant's history of genital ulcers and education levels) were significantly associated with HSV-2 seroprevalence. Our study found that only 6% (11/169) of the participants reported having ever consumed alcohol. This is in contrast to another recent study from the same region reporting alcohol consumption among adolescents and youth around 70% [30]. The inconsistency could be due to the differences in populations as the previous studies focused on adolescents and youth students with unknown HIV serostatus, presumably due to continuous health education provided at CCFCC.

Our study was conducted in an HIV-1 clinic setting with a relatively limited number of registered individuals. Information on participant's sexual behaviour and other risk behaviours were collected through interviews, an approach known to be prone to under-reporting [31,32]. With these limitations, our findings need to be interpreted with caution. Further studies delineating the risk factors for adolescents and youth contracting HIV-1 and HSV-2 infections are necessary to help inform prevention strategies.

## Conclusions

Our findings suggest that HSV-2 seroprevalence increased by age among adolescents and youth living with HIV-1. Age-specific approaches might play an important role in interventions targeting HSV-2 infection.

## Acknowledgements

We would like to thank the study participants and their parents/guardians for consenting and participating in the study. Staff members of the Child-Centred Family Care Clinic (KCMC) and Kilimanjaro Christian Research Institute (KCRI) are thanked for their help in the data collection and laboratory experiments, respectively.

## Author's contributions

RM, IK, ZT and TLK conceived and designed the study. RM and AN collected the samples and conducted laboratory experiments. RM and FF analyzed the data. RM drafted the manuscript with contributions from IK, ZT, VB, and TLK. All the authors read and approved the final version of the manuscript.

## Funding

This study received financial support from Copenhagen University Hospital, Rigshospitalet, Denmark.

## Ethical approval and consent to participate

The study was approved by the Kilimanjaro Christian Medical University College (KCMUCo) Research and Ethics

Review Committee (approval number 2026 of 2017). Participants were explained about the study before sample collections. All participants aged 18 years and above provided signed informed consent before enrolled in the study. For those below 18 years, consent was sought from their parents or guardians and assent was obtained from the minors before enrolment into the study.

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**Conflict of interests:** No conflict of interests is declared.