

# Factors Associated with Tungiasis Infestation among School age Children in Ugenya Sub-County, Siaya County, Kenya

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# Summary

#### BACKGROUND

*Tungiasis* recognized as a neglected parasitic tropical disease by the WHO is caused by female *Tunga penetrans* (jigger, Flea) which has remained a significant public health problem. It affects resource-poor communities causing various health disabilities. By 2016, about 2.6 million Kenyans were infested out of whom 1.5 million were school going children who developed physical and mental disabilities.

#### **OBJECTIVE**

The study aimed at assessing factors that influence practices towards prevention and control of *tungiasis* infestation among school children in Ugenya Sub-County, Kenya.

#### METHODOLOGY

A descriptive cross-sectional study design and quantitative data collection method was used between January and March 2018. A total of 385 participants clinically examined for the presence of *tungiasis* after consenting were enrolled. Male 200 (51%) and female 185(49%). Using standard methods, macroscopic examination was performed by carefully inspecting the legs, feet, hands and arms of school pupils aged 6 to 14 years. Initially, information meetings were held with County officials in the line ministries, School committees and Community representatives to sensitize on disease prevention, control, and ownership of the process. Community Health Workers (CHWs) getting involved on how to identify classification of *tungiasis* as performed for them and by field officers. Simple random sampling technique was applied for selection. A pretested semi-structured questionnaire had been administered in English/ Vernacular on approximately 10% of the participants. Data was keyed-into Microsoft Excel and analyzed using IBM SPSS version 23.

#### RESULTS

The overall prevalence was found to be 31.1% which was slightly higher than a previous community-based study that had a prevalence of 25% [24] indicating higher infestation in schools than in the community. Remarkably, the majority of pupils infested in classes 5-6 were 191(49%). Gender of pupils (p<0.005) showed a statistical significance with *tungiasis* infestation. Infestation of family member at (p<0.005) and action taken when family member was infested (p=0.042) with posted level of significance. A positive relationship between sleeping area in the house (p=0.048) as well as waste disposal (p=0.017) with *tungiasis* infestation.



#### **CONCLUSIONS**

Factors including gender, invasion of a family member, action taken upon infestation, sleeping area in the house and waste disposal, significantly predicted *tungiasis* infestation. Diagnosis of *tungiasis* having been done by experienced community members and confirmed using 'Fortaleza classification'. following surgical extraction of the flea, thorough cleansing and covering of the remaining crater with a topical antibiotic cream to prevent secondary infection was guaranteed. [2] School age children in developing countries bore the greatest health burden from neglected tropical infections including *tungiasis*.

#### RECOMMENDATIONS

The key word here is hygiene. Imperative control programs to adopt a more comprehensive approach including School and Community health education to reduce the spread and morbidity from *tungiasis* is essential. However, sustainable control measures against *tungiasis* could only be developed if the epidemiological situation is well understood [4] Oral antibiotics and un-expired Tetanus prophylaxis should be readily availed within such endemic areas.

Key words: Practice, *Tungiasis*, Prevention, Control, Ugenya, Infestation

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### Introduction

*Tungiasis* recognized as a neglected tropical disease by the World Health Organization is also referred to as *tungiasis* infestation due to the fact that it typically occurs in populations living in poverty with poor housing, inadequate sanitation and in close contact with domestic animals, akin to other neglected diseases. [1].

The disease affects communities living in precarious conditions trapping them in a vicious cycle of poverty and disease [2]. It was endemic in tropical and subtropical countries [3]. In the previous years, the public health significance of *tungiasis* in resource-poor populations had been highlighted from different countries, including Brazil, Argentina, Haiti, Nigeria and Tanzania. It was an exclusively tropical *ectoparasitosis* that occurred in South America, the Caribbean, and Sub-Saharan Africa [4].

School age children in developing countries bore the greatest health burden from neglected tropical infections including *tungiasis*. Accounting for an estimated 20% of the disability-adjusted life years lost due to infectious diseases in children less than 14 years old [5].

About 2.6 million Kenyans were infested with *tungiasis* out of whom 1.5 million were school going children who were physically and mentally disabled [6].

The impact of the disease was felt heavily among poverty stricken rural-agricultural communities and was believed to inhibit progress and development. Serious infestations could lead to severe inflammation leading to loss of toenails, auto amputation of digits and death may also occur The risk of secondary infection such as tetanus was also high. The *tungiasis* menace deprived pupils concentration in class, affected extra curriculum participation often leaving them lethargic and slow in action. Adults who had been infested by tungiasis were completely dependent as they could not actively take part in day to day activities [7 - 8].

In Ugenya Sub-county it was believed that *tungiasis* infestation was a curse and once infested, the individual were discriminated. In north-east Brazil, there was considerable seasonal fluctuation of the attack rate [9]. It had been observed that during the rainy seasons, the incidence of new lesions decreases significantly while during dry month's new cases of tungiasis showed up every day [10].

Given considerations were required as far as Tungiasis treatment approaches were concerned. A number of surgical treatment methods were availed. The flea could be removed from its cavity with sterile instruments, but that was more difficult when the flea was engorged [9]. The orifice was to be enlarged and



the entire nodule curetted or excised. Following surgical extraction of the flea, thorough cleansing and covering of the remaining crater with a topical antibiotic cream to prevent secondary infection was important. [2] A course of oral antibiotics could be instituted on suspicion of secondary infection. Tetanus prophylaxis should readily available.

*Tungiasis* had a focalized distribution with the disease occurring predominantly in impoverished populations. According to pilot investigations, *tungiasis* was highly prevalent in parts of Coast, Nyanza, Rift Valley, Western and Central regions of Kenya. However, sustainable control measures against *tungiasis* could only be developed if the epidemiological situation was well understood [4]. Children were the hardest hit with a preceding study of 2014 estimating a point prevalence of 57% among children aged 5-12 years in Murang'a North and South districts, Kenya [5], [11].

Factors that have been reported to aggravate the problem include earthen and dusty floors, non-regular use of closed footwear, common resting place outside the house and pigs on the compound [12]. Sleeping on the floor and crowded sleeping areas have also been cited [13]. Others are living with animals in the same house [6], occurrence of animals like dogs, cats, rats, cows, goats, chicken in the compound [6], [14] and low level of education [15].

Unhygienic conditions have also been identified as the major causes of *tungiasis* in Kenya [16]. Though such information is already available, more work on epidemiology especially in vulnerable age groups such as children and the aged in other endemic regions need to be pursued. Control measures for *tungiasis* can only be developed if epidemiological situation is well understood. To fill this gap and validate anecdotal observations, a cross-sectional study was carried in Ugenya sub-county, which provided evidence-based information and form basis for further studies and effective sustainable control measures among school age children and the community as a whole in Kenya.

# Methodology Study Site

This study was conducted in Ugenya Subcounty in Nyanza region of Kenya. It was one of six constituencies of Siaya County which had a child rich population, whereby 0-14 years old constitute 45% of the total population. However, the county was at the onset of a fertility decline as 42.9% of households had 0-3 household members and 39.4% of households had 4-6 members.

In Siaya County, 11% of the residents with no formal education were working for pay, 13% of those with primary education and 22% of those with secondary or above level of education. Work for pay for those with secondary or above level of education was highest in Nairobi at 49% which was twice the level in Siaya. Siaya County had a population of 993,183 [17]. It had four wards namely; West Ugenya, East Ugenya, Ukwala and North Ugenya.

The rural setup covered an area of 310.20 square kilometers with. improved sources of water comprising of protected spring, protected well, borehole piped into dwelling, piped and rain water collection. Unimproved sources included pond, dam, lake, stream/river, unprotected spring, unprotected well, jabia, water vendor and others. In this County, 36% of residents used improved sources of water, with the rest relying on unimproved sources. A total of 49% of residents used improved sanitation, while the rest used unimproved sanitation. Statistics from the Ministry of Health, Siaya county in 2012 showed that over 200,000 people in the county were infested with *tungiasis* [18].

# Study Design, Study Population, Sample size and Sampling procedure

This was a school based descriptive crosssectional study that took place between January and March 2018, a dry season when prevalence of *Tungiasis* was known to be at its peak. The study adapted both qualitative and quantitative data collection approaches. Studies by [10] and [19], which had indicated that School age children aged 6 to 14 years were the most affected.

# Sample Size Determination

The sample size was determined using the Daniel, 1999 formula which uses prevalence as the P value [20]. The prevalence of *tungiasis* ranges between 15% to 60% and therefore a proportion of 50% was used to draw the sample size[21].



To calculate the adequate sample size, the following formula indicated for prevalence study was applied:

 $n = Z^2 P(1-P)/e2 [22],$ 

where: n was a minimal required sample size,

z a standard normal deviation for

a 95% confidence interval,

p an estimated prevalence of *tungiasis*, and

e the degree of precision.

The prevalence was estimated by random at 50% since no previous study on *tungiasis* had been done in Ugenya sub county.

> Thus, for: z = 1.96, p = 0.5, and

$$e = 0.05,$$

n was estimated at 385 children.

Hence, the calculated sample size was 385.

# **Data Collection**

Before the onset of the study, information meetings were held with County officials in the line ministries, School committees and Communities representatives to sensitize on the disease prevention and control including ownership of the process before it commenced. Community Health Workers (CHWs) were involved basically from diagnosis including a pre-examination on how to identify classification of *tungiasis* performed for them and the field officers. A pretested semi-structured questionnaire was conducted among 385 participants and administered in English, or the local language to the participants.

For the school-based survey; interviewers administered semi - structured questionnaires which were developed and used as one of the data collection tools to elicit information on the demographic data such as; socio-demographic factors including; sex, age, and education. Knowledge and practices related to *tungiasis* as well. for example:

- 1. Knowledge on transmission.
- 2. Regular use of footwear.
- 3. Common resting place.
- 4. Preventive measures.
- 5. Treatment.

Before administration, approximately 10% of the 385 questionnaires (38 questionnaires) were pretested by administering them to schools from an area neighboring the survey site. Revision after pretesting was done before implementation (Appendix 1).

## Quantitative Data Management and Analysis Data analysis

Data was entered into Microsoft Excel version 2016 spreadsheet for cleaning, transferred to Statistical Package for Social Sciences (IBM SPSS) Version 23 for transformation and exploratory data analysis. Data analysis involved descriptive statistics (means, standard deviations, medians and frequency distributions as well as computation of Prevalence Rate Ratios (PRR). Chi square tests were used for the associations between variables where applicable. Results were presented in frequency distribution tables and charts. Differences between the parameters of estimates was deemed statistically significant at p < 0.005. All variables that remained in the model are presented.

## Validity and reliability of Instruments

Validity of the instrument was ensured by pretesting the questionnaires in four schools to check for proper recording of responses, completeness and if the results were the same and reflected the variables under study. The researcher also ensured that the results were not influenced by any biases, interest or perspectives. Internal consistency reliability test was conducted using Cronbach Alpha Test, the standard minimum value of

*a* = 0.7 [23].

# Results Demographics

The 385 respondents' class distribution with pupils from classes 5-6 being the majority at 191(49%) while those in class 7-8 were 189 (48%) and respondents who were in class 3-4 at (3%) the least in the category.

The participants who had ever been infested with *tungiasis* were 333 (85.4%). The respondent's gender distribution was, 200 (51%) male and 185(49%) female. There was a significant association between gender and *tungiasis* infestation at ;

- $(\chi 2 = 4.383,$
- df = 1,
- p = 0.036).

The age of 10 - 15 years category was the most dominant at 306 (78.3%) while 15 - 18 years category was at 81 (21.0%). Christians were the majority of respondent's parents at 383(99.2%) (*Table 1*).



### Table 1: Socio-Demographic Characteristics among Pupils Sampled from Different Schools in Ugenya Sub-County, Siaya County in Kenya

Socio-Demographic Characteristics		Tungiasis	Infestation		$\chi^2$	df	P-value	
		No n (%)	Yes n (%)	Total				
Respondents class	spondents 3 - 4		7(70.0)	10	4.177	2	0.124	
	5 - 6	22(11.5)	164(88.5)	186				
	7 - 8	32(16.9)	152(83.1)	184				
Sex	Male	22(10.9)	178(89.1)	200 4.383		1	0.036	
	Female	35(18.4)	150(81.6)	185				
Age	5 - 9	0(0.0)	2(100.0)	2	1.036	3	0.793	
	10 - 15	47(15.4)	254(84.6)	301				
	15 - 18	10(12.2)	71(87.8)	81				
	19 and Above	0(0.0)	1(100.0)	1				
Family size	1 - 3	4(17.4)	19(82.6)	23	2.550	3	0.466	
	4 - 7	36(15.5)	196 (84.5)	232				
	9 - 11	15(12.4)	105(87.6)	120				
	12 and Above	3(30.0)	7(70.0)	10				
Household head	Business	10(20.5)	50(79.5)	60 44.729		28	0.023	
occupation	Farmer	22(12.4)	155(87.6)	177				
	Formal Employment	5(5.3)	54(94.7)	59				
	Not working	2(20.0)	8(80.0)	10				
	Informal employment	9(15.4)	70(84.6)	79				
Religion of the parent	Christian	58(15.1)	325(84.9)	383	0.535	1	0.465	
	Muslim							



# **Diagnosis and Case Definition**

Macroscopic examination was performed by carefully inspecting the legs, feet, hands and arms. To guarantee privacy, other regions of the body were not examined. This approach was considered as acceptable because in endemic communities, more than 99% of *tungiasis* lesion occur on these areas [6].

Diagnosis of *tungiasis* was done by experienced community members and confirmed using 'Fortaleza classification'[19] whereby the following symptoms are considered to be pathognomonic for *tungiasis*:

- A red-brown itching spot with a diameter of 1-2mm (early stage),
- A yellow-white watch glass-like patch with a diameter of 3-10mm with a central dark spot (mature stage).
- A brown-black crust with or without surrounding necrosis (dead flea=last stage)
- Embedded sand fleas with evidence of

manipulation by the patient or his/her caretaker with instruments such as needles or thorns.

To reduce observational bias, visual examinations and interviews were conducted by different investigators.

## Prevalence

On the number of pupils who had *tungiasis* per class in a sample of 10 pupils 4 (10.5%) indicated that 0, 1 or 6 which is 0%, 10% and 60% prevalence respectively.

9 (23.7%) of the respondents indicated that 2 of the pupils had *tungiasis* which was 20% prevalence.

The respondents which indicated that 3 and 4 had *tungiasis* which is 30% and 40% prevalence respectively per class.

6 (15.8%), the respondents which indicated 5 pupils had *tungiasis* which translates to 50% *tungiasis* prevalence per class. 3 (7.9%) and 1 (2.6%) of the respondents indicated that 7 or 8 pupils had *tungiasis* per class which is 70% and 80% *tungiasis* prevalence per class respectively. Averagely there was 118 (31.1%) *tungiasis* prevalence (*Table 2*).



			No. of pupils with <i>tungiasis</i> per class Sample of 10 pupils Yes										
		0	1	2	3	4	5	6	7	8	- Total		
School	Ligala	Count	1	0	2	0	1	0	0	0	0	4	40
Name		School Name	25.0%	0.0%	50.0%	0.0%	25.0%	0.0%	0.0%	0.0%	0.0%	100.0%	
	Siwar	Count	0	0	1	1	0	0	1	0	1	4	40
		School Name	0.0%	0.0%	25.0%	25.0%	0.0%	0.0%	25.0%	0.0%	25.0%	100.0%	
	Random	Count	0	0	1	0	0	0	0	0	0	1	10
		School Name	0.0%	0.0%	100.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	100.0%	
	Uring	Count	1	1	0	1	0	0	0	0	0	3	30
	School Name	33.3%	33.3%	0.0%	33.3%	0.0%	0.0%	0.0%	0.0%	0.0%	100.0%		
	Nyaharwa	Count	0	1	0	1	1	0	0	0	0	3	30
		School Name	0.0%	33.3%	0.0%	33.3%	33.3%	0.0%	0.0%	0.0%	0.0%	100.0%	
	Ng'ang'a	Count % within School Name	0	0	1	0	1	0	1	0	0	3	35
			0.0%	0.0%	35.5%	0.0%	35.5%	0.0%	35.3%	0.0%	0.0%	100.0%	
	Lwero	Count % within School Name	0	2	1	0	0	1	0	0	0	4	40
			0.0%	50.0%	25.0%	0.0%	0.0%	25.0%	0.0%	0.0%	0.0%	100.0%	
	Nyalenda	Count % within School Name	0	0	2	1	0	1	0	0	0	4	40
			0.0%	0.0%	50.0%	25.0%	0.0%	25.0%	0.0%	0.0%	0.0%	100.0%	
	Ukela	Count % within School Name	0	0	0	1	0	0	2	1	0	4	40
			0.0%	0.0%	0.0%	25.0%	0.0%	0.0%	50.0%	25.0%	0.0%	100.0%	
	Siria	Count % within	0	0	0	1	2	1	0	0	0	4	40
		School Name	0.0%	0.0%	0.0%	25.0%	50.0%	25.0%	0.0%	0.0%	0.0%	100.0%	
	Ndenga	Count % within	2	0	1	0	1	0	0	0	0	4	40
		School Name	50.0%	0.0%	25.0%	0.0%	25.0%	0.0%	0.0%	0.0%	0.0%	100.0%	
Total		Count % within	4	4	9	6	6	4	4	1	1	38	385
		School Name	10.5%	10.5%	23.7%	15.8%	15.8%	7.9%	10.5%	2.6%	2.6%	100.0%	
			0	. 4	18	. 18	. 24	. 15	5 24	I 7	. 8	. 118	31.1%

Table 2: Prevalence of Tungiasis in different Schools in Ugenya Sub county, Siaya County in Kenya Crosstab



# **Practices towards** *Tungiasis* **Infestation**

*Table 3* shows the output of the ANOVA analysis and whether there was a statistically significant difference between the group means. The model was significant at the value p<0.001, which was below 0.005 level of significant and therefore, there was a

statistically significant difference in the mean *tungiasis* infestation. The model had statistically significant predictor variables influencing it. Sleeping place at ( $\beta$ =-0.101, t=-1.994, p=0.048), any member of family that was infested by *tungiasis* at ( $\beta$ =-0.101, t=3.011, P=0.003), action taken when the member of the family had *tungiasis* at ( $\beta$ =-0.030, t=-2.043, P=0.042) and the disposal place of waste product at home at ( $\beta$ =-0.057, t= -2.417, P=0.017).

 Table 3: Practices Contributing towards Tungiasis Infestation among Pupils in Sampled Schools in Ugenya Sub - County, Siaya County, Kenya

Coefficients <sup>a</sup>									
Model	Unstand Coeffi	lardized cients	Standardized Coefficients	t	Sig.				
	B Std. Error		Beta						
1 (Constant)	1.180	158		7.485	.000				
The sleeping place	028	.014	138	-1.994	.048				
Member of family that is infested by <i>tungiasis</i>	.101	.034	.208	3.011	.003				
Action taken when the member of the family has <i>tungiasis</i>	030	.015	144	-2.043	.042				
<i>Tungiasis</i> removal	003	.010	021	298	.766				
Knowledge of harm associated with physical removal of <i>tungiasis</i>	.016	.035	.031	.443	.658				
Application of products and type	025	.023	070	-1.062	.289				
The disposal place of waste product at home	057	.024	167	-2.417	.017				
Burning of waste	.084	.049	.116	1.727	.086				
Wearing of shoes to school	059	.063	086	947	.345				
Frequency of wearing shoes to school	.038	.024	.160	1.608	.109				
The type of shoes one wears	030	.023	110	-1.328	.186				
Frequency of washing uniforms	.035	.019	.123	1.830	.069				
Part of house one sleeps	001	.020	005	068	.946				
The type of floor in the house	024	.025	066	972	.332				

<sup>a.</sup> Dependent Variable: Infested with *tungiasis* 



# Discussion

This study found a point prevalence of *tungiasis* at 31.1% which was slightly higher than what was reported in an earlier community-based study in Kenya that had a prevalence of 25% [24]. This possibly indicated a higher *Tunga* penetrans' infestation in schools than in the community. It was however lower than what was reported in other countries like Nigeria 45.2% and Tanzania 42.5% [25], it could be attributed to various confounding factors including low levels of sanitation.

Study findings indicate a significant association between gender of pupils and *tungiasis* infestation. The data difference is attributed to exposure and environmental factors including socio-economic and cultural aspects, rather than a difference in susceptibility. Thus, we speculate that gender differences were related to different exposure and disease-related behavior [10].

These findings have been consistent across studies but levels of infestation in each gender seem to differ from community to another. Research in Cameroon and Trinidad found statistically significant differences between gender with which males carried higher disease burden compared to females [7], [26], this could be due to the fact that hygiene levels among males are lower as compared to females.

Daily chores among boys could expose them to the *ectoparasite* while less wearing of shoes than in girls renders them more vulnerable to jigger infestation. Higher prevalence was noted in children between classes 5-6 in the this study. The significance of the grade variables in this study is consistent with a previous study in rural western Tanzania [25] which reported a high prevalence of *tungiasis* in children of this age group. In addition, high prevalence between ages 5–14 year can be attributed to the fact that they are more exposed due to them playing barefoot [4] and also getting re-infected at school especially in endemic areas.

Current study findings post significance on infestation and action taken when family member was infested. Previous studies have indicated that perception of family and community members towards *tungiasis* was critical since it influences their reaction toward *tungiasis* prevention and control [27]. It is therefore important to inculcate the right perception in household members in order to develop a positive behavior change for sustainable *tungiasis* prevention and control among household members. However, social efforts to improve hygiene, welfare and standard of living do provide additional protection against the *tungiasis* flea. In addition, *tungiasis* can be regarded as a neglected poverty-associated disease [28].

Further analysis on this study displays the association between disposal of waste products at home and *tungiasis* infestation. It is speculated that dirty and poorly maintained environment is a preferred breeding site for the flea, as there is abundant organic material for the larvae to feed on. The findings concur with a study by [29] which reported that improving sanitation and waste collection reduced the incidence of *tungiasis*.

Home hygiene is important in order to control pests and provide pleasant atmosphere to the household members [7]. These findings do also concur with [27] where they realized that maintenance of personal hygiene, improvement of dwelling place, health education and maintenance of hygiene in the surrounding environments were suggested measures to tackle *tungiasis*. However, the effectiveness of these measures is difficult to predict and they are more costly than cementing floors of houses, confining pigs to pigpens and realizing health education [7].

Further findings on inferential analysis show a positive significance on place of sleep within the house and *tungiasis* infestation. It concurs with a study by [8] which reported other modifiable risk factors as the resting places within the house and that when humans live in close contact with infested animals, the risk of infestation is high [8]. Almost half of the respondents in the study kept domestic animals in the main house and this poses a risk factor to *tungiasis* infestation.

Intervention strategies would be greatly enhanced if spraying or dusting of houses with insecticides is integrated in mass control programs. Taken together, it seems plausible that the common feature here was that, transmission took place where respondents spend most of their time since the eggs must fall onto the ground complete their life-cycle and get back to the human host. If this is true then homes and schools should be looked at as possible venues of transmission and be investigated further in longitudinal studies.



Being a cross sectional study, the aspect of cause and effect could not be investigated. This is because cross sectional studies are only able to establish an association or a relationship between two or more variables. For a study to claim causality which means that changes in one variable measured directly caused changes in another, a longitudinal study has to be carried out. Future studies can attempt to fill this gap by conducting longitudinal studies on variables of interest.

# Conclusion

Study concludes that gender of pupils, infestation and action taken when family member was infested with *tungiasis* as well as disposal place of waste significantly influence *tungiasis* infestation. These factors influence infestation levels, prevention and control of *tungiasis* infestation both at school and at home. Control programs need to adopt a more comprehensive approach including School and Community health education which is imperative to significantly reduce the spread and morbidity from *tungiasis*. Sustainable control measures should include improving sanitation and hygienic behaviors among school age children and host communities and the incorporation of its control in the school health programmes in Kenya.

# List of Abbreviations

- CPHR:Center for Public Health ResearchESACIPAC:Eastern and Southern Africa Centre of<br/>International Parasite ControlSERU:Scientific & Ethics Review Unit
- SPSS : Statistical Package for Social Sciences
- **IBM :** International Business Machines
- **CHW :** Community Health Workers

## Declarations

# Ethics Approval and Consent to Participate

This study was approved by the KEMRI Scientific Ethical and Review Unit (SERU) protocol No. (KEMRI/SERU/ESACIPAC/P003/3531).

The study used questionnaires uniquely coded with results of each questionnaire being kept in strict confidence. Participating in the study was voluntary and one could withdraw at any point. The purpose of the study and its objectives were explained to local authorities, opinion leaders, headteachers, and community members. Informed consent and assent was obtained from the participating respondents in writing. Written parental consent was obtained for participants under 16. Subjects were assured about confidentiality of information obtained from them and personal identifiers were removed from the data set before analysis.

#### **Consent to Publish**

Not applicable

# Availability of Data and Materials

That all data used in the manuscript is available for sharing; including all relevant raw data, will be freely available to any scientist wishing to use them for noncommercial purposes, without breaching participant confidentiality.

# **Competing Interests**

The authors declare that they have no competing interests.

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# **Authors' Contributions**

- JM conceived of the study, participated in its design coordination and helped to draft the manuscript.
- EG participated in the design of the study and helped to draft the manuscript.
- MHA- Participated in data analysis and draft manuscript preparation.

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