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2

3 **TITLE:** Risk factors for bruli ulcer in a referral mission hospital in anambra state,  
4 Nigeria: A case control study

5

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24 **Short Running Title:** Buruli ulcer risk factors

25

26 **Guarantor of Submission:** The corresponding author is the guarantor of  
27 submission.

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33 **ABSTRACT**

34

35 **Aims**

36 Buruli ulcer is a chronic debilitating skin disease believed to be caused by an  
37 environmental bacterium – *Mycobacterium ulcerans*. Its mode of transmission is still  
38 elusive. Buruli ulcer can cause permanent disability and deformity which may  
39 severely limit a person's ability to carry out normal daily activities. The associated  
40 stigma may greatly restrict the social participation by affected persons. Though early  
41 detection and adequate medical and surgical treatment can minimize future disability  
42 and complication, recognizing the risk factors of this disease can lead to adoption of  
43 appropriate preventive strategies.

44

45 **Methods**

46 A case-control study of 120 patients (40 cases and 80 matched controls) was  
47 undertaken in a referral mission hospital. A structured questionnaire was used to  
48 collect data from subjects. The folders of case subjects were also reviewed.

49

50 **Results**

51 Significantly higher proportion of Buruli ulcer cases were males when compared to  
52 the control group ( $\chi^2 = 3.84$   $p = 0.05$ , OR = 2.22). Poor education ( $\chi^2 = 14.27$ ,  
53  $p=0.0003$  and OR 5.13), visit to water bodies ( $\chi^2 = 67.78$ ,  $p=0.00000$  and OR = 63),  
54 No BCG vaccination ( $\chi^2 = 6.79$ ,  $p=0.01$  and OR = 2.79), pet in the house ( $\chi^2 = 9.25$ ,  
55  $p=0.002$  and OR = 3.55), poor drinking water source ( $\chi^2 = 45.32$ ,  $p=0.00000$  and OR  
56 = 19.52), no preventive measure ( $\chi^2 = 18.3$ ,  $p=0.0005$  and OR = 6.27) were  
57 significantly associated with Buruli ulcer.

58

59 **Conclusion**

60 This study identified, regular visit to water bodies, male gender, poor education, lack  
61 of BCG vaccination and poor drinking water source as risk factors for Buruli ulcer, in  
62 the face of apparent lack of awareness of disease and its risk factors among the  
63 subjects. Aggressive public enlightenment is recommended to hopefully reduce the  
64 prevalence of Buruli ulcer disease.

65 **Keywords:** Buruli ulcer; Risk factors, Nigeria

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EARLY VIEW

97 **INTRODUCTION**

98 Buruli ulcer is a chronic debilitating skin disease caused by mycobacterium ulcerans.  
99 It is an acid-fast bacillus and belongs to the same family of organisms that cause  
100 tuberculosis and leprosy. The mode of mycobacterium ulcerans transmission is not  
101 fully understood although the disease is known to be linked to contaminated water.  
102 Areas affected by buruli ulcer disease are located near stagnant or slow moving  
103 waters.

104 Buruli ulcer is one of neglected tropical diseases and has been observed more  
105 frequently during the rainy season in Africa and exposure may occur in muddy  
106 farming fields. Transmission of the organism is probably via skin trauma [1,2].  
107 Insects may also play a role in some cases, though not in all foci where transmission  
108 occurs. Water insects (Naucoris and Belostomaspp) have been implicated in  
109 laboratory transmission of infection but their potential role as vectors has been  
110 questioned by studies in west Africa [3]. In Australia, salt marsh mosquitoes appear  
111 to have positive polymerase chain reaction signals [4]. Intermediate hosts including  
112 aquatic animals may also play a role. Amoebae have also been implicated but their  
113 role in transmission is limited. Human to human transmission is rare.

114 The disease manifests itself as papule, nodules, plaques, ulcers and oedematous  
115 areas of the skin. Buruli ulcer often starts as a painless swelling (nodules). It can  
116 also initially present as a large painless area of induration (plague) or a diffuse  
117 painless swelling of the legs, arms or face (oedema) [5]. The local  
118 immunosuppressive properties of the mycolactone toxin enable the disease to  
119 progress with no pain and fever. Without treatment or sometimes during antibiotic  
120 treatment the nodules, plaques or oedema will ulcerate within four weeks with the  
121 classical undermined borders. Occasionally bones are affected causing gross  
122 deformities [5]. The distinctive features of a buruli ulcer include- undermining edges,  
123 white cotton wool-like appearance and thickening and darkening of the skin  
124 surrounding the lesion [5].

125 In a known endemic area, diagnosis of buruli ulcer can be made based on clinical  
126 observation that considers clinico-epidemiological features which indicate that most  
127 cases are in children under 15years of age, about 85% of lesions are on the limbs  
128 and lower limb lesions are twice as common as upper limb lesions. In addition to the

129 clinical diagnosis, at least one of the following laboratory findings is required to  
130 confirm the diagnosis of buruli ulcer: acid-fast bacilli (AFB) in a smear stained by the  
131 Ziehl-Neelsen technique, histo-pathological study of a biopsy specimen showing  
132 typical necrosis and acid-fast bacilli, positive polymerase chain reaction (PCR) test  
133 for mycobacterium ulcerans and positive culture of mycobacterium ulcerans [6].

134 In spite of the seeming high prevalence of buruli ulcer, the disease remains generally  
135 poorly understood. Diagnosis is largely presumptive and the associated risk factors  
136 poorly defined. This study is aimed at identifying possible risk factors for buruli ulcer  
137 in the study area. It also determines the knowledge of buruli ulcer among the  
138 respondents and explores preventive measures adopted by the affected subjects.

139

## 140 **MATERIALS AND METHODS**

141

### 142 **Study design and case definition**

143 A case-control study was designed in a mission hospital located in Anambra state,  
144 Nigeria and classified as a secondary healthcare facility. It is a referral centre for  
145 buruli ulcer disease and is largely supported by German Leprosy and Tuberculosis  
146 Relief Association.

147

### 148 **Study Population**

149 The case study population consists of patients diagnosed with Buruli ulcer. All the  
150 patents with suspected buruli ulcer that were referred to the centre and examined by  
151 the Physician in charge of buruli ulcer disease and confirmed by laboratory tests  
152 using Ziehl-Neelsen technique and polymerase chain reaction (PCR) test. The  
153 control population consists of persons not diagnosed of buruli ulcer disease or  
154 similar illness but who share similar socio-demographic and geographical  
155 characteristics as the cases. The control population was selected to be of the same  
156 sex and age range of  $\pm 2$  SD with the case (study) population.

157

### 158 **Selection Criteria for Subjects**

159 Inclusion criteria for case: 1) Patients diagnosed with Buruli ulcer and have been  
160 confirmed by polymerase chain reaction.

161 Exclusion criteria: Those whose ulcer have not been confirmed.

162

### 163 **Sample Size Determination**

$$164 \quad X = \frac{U\sqrt{[\pi_1(1 - \pi_1) + \pi_2(1 - \pi_2) + v\sqrt{[\pi(1 - \pi)]}}$$

$$165 \quad (\pi_1 - \pi_2)^2$$

166  $U = 1.28$  (where power is 90%).

167  $V = 1.96$  (where significance level = 5%).

168  $\pi_1$  = Proportion of control exposed.

169  $\pi_2$  = Proportion of cases exposed.

170 It can also be calculated from,

$$171 \quad \pi_2 = \frac{\pi_1 OR}{1 + \pi_1(OR - 1)}$$

171

172 Where OR = Odds ratio;  $\pi_1 = 0.1$ ;  $\pi_2 = 0.25$

173  $X = 57.30 \approx 60$ ; Minimum sample size =  $2X = 120$ .

174

### 175 **Sampling Method**

176 A convenience sampling method was used to collect data for the calculated sample  
177 size. The folders of 40 patients diagnosed of Buruli ulcer were assessed through the  
178 hospital register. Using the folder numbers, the patients' records were traced and  
179 vital information required for the research was collected. The patients were also  
180 identified for primary data collection using interviewer administered questionnaires.

181 The control groups 80 subjects consisted of apparently normal persons of similar  
182 sex, age and geographical location attending the hospital or live within the region  
183 where the patients reside. The same questionnaires were administered on the  
184 control.

185

186

187

188 **Tool for Data Collection**

189 The data was collected using an interviewer-administered questionnaire. This was  
190 used to collect data on socio-demographic characteristics, other relevant risk factors  
191 as well as knowledge of Buruli ulcer in both the case and the control groups.

192

193 **Data Analysis**

194 The data obtained was analyzed using the Microsoft Excel of the computer.  
195 Descriptive summary statistics such as mean were derived and data was presented  
196 in tables. Odds ratio was calculated for the risk factor under evaluation. Association  
197 between the variable were calculated using appropriate statistical test and the level  
198 of significance set at  $p \leq 0.05$ .

199

200 **Limitations**

201 Some limitations were encountered in the course of the study and they include: Little  
202 research has been conducted on Buruli ulcer in the study area, with resultant paucity  
203 of literature on the subject matter.

204

205 **Ethical Consideration**

206 Ethical approval for the project was obtained from the NAUTH Ethics Committee  
207 through the office of the Head of Department, Community Medicine and PHC,  
208 Nnamdi Azikwe University Teaching Hospital, Nnewi, Nigeria. Appropriate  
209 permission was also obtained from the Manager of the Mission hospital and heads of  
210 various sections of the records department. The nature of the study was clearly  
211 explained to them before obtaining the approval. Adequate confidentiality was  
212 maintained during the use of the patient's folder. Verbal informed consents were  
213 obtained from the subjects.

214

215 **RESULTS**

216 A total of 120 subjects of 40 cases of Buruli ulcer and 80 controls were interviewed.  
217 The study population (cases) and control groups were very similar in terms of age  
218 ( $P > 0.05$ ). Hence, the case and control groups were age-matched as shown in table 1  
219 above.



220 In table 2, the distribution of the subjects' educational levels, occupations, social  
221 history and co-morbidities are displayed.

222 Table 3: Shows the distribution of respondents according to other socio-  
223 demographic characteristics: Sewage disposal method, sources of drinking water,  
224 type of living house, refuse disposal method, pet in the house, BCG vaccination,  
225 BCG scar present and Visit to water body.

226 Table 4 depicts the distribution of the cases and control (where applicable) according  
227 to awareness, characteristics and management of the ulcer among them. It shows  
228 that 5% and 1.25% of cases and control have heard about BU, just as the knowledge  
229 of risk factors in both groups was also poor. Lesion was found mainly in the lower  
230 limbs (70% of cases). Fifty per cent of cases applied no preventive measure while  
231 40% of the control adopted the use of ITN as a preventive strategy. About 97% of  
232 BU victims applied wound dressing at a local patent medicine vendor's, while 95% of  
233 them took herbal concoctions before seeking help in hospital.

234 Table 5 shows that significantly higher proportion of BU cases than the control were  
235 males ( $X^2=3.84$ ,  $p=0.05$ ). The odd of BU occurring among male patients is about 2  
236 times more than would occur among the females (OR = 2.22). Similarly, poor  
237 education, visit to water bodies and absence of BCG vaccination are significantly  
238 associated with BU ( $X^2 =14.27$ ,  $P=0.005$ ;  $X^2 =67.78$ ,  $P=0.00001$ ,  $X^2 =6.79$ ,  $P=0.01$ ,  
239 respectively). The chances Of BU occurring among subjects exposed to these risk  
240 factors are about 63, 5 and 3 times more when compared with the likelihood among  
241 those without these conditions, (OR =63 visit to water bodies; OR =5.13 poor  
242 education; OR =2.79 absence of BCG vaccination respectively). In the same vein,  
243 presence of pet in the house, obtaining drinking water from body water sources and  
244 failure to adopt any preventive measure are markedly associated with BU ( $X^2=9.25$ ,  
245  $P=0.002$ , OR=3.55,  $X^2=45.32$   $P=0.00000$ , OR=19.52  $X^2=18.30$ ,  $p=0.00005$ ,  
246 OR=6.27). The corresponding odd's ratios for these risk factors range from 2.22 to  
247 63. However, farming, open dumping of refuse, poor sewage disposal are not  
248 significantly associated with BU( $X^2 =3.20$ ,  $p=0.07$ , OR =2.15,  $X^2=0.17$ ,  $P=0.83$ ,  
249 OR=1.19,  $X^2=1.49$ ,  $P=0.15$ , OR=1.71 etc respectively).

250

251

252 **DISCUSSION**

253 This study analyzed some common risk factors associated with Buruli ulcer. The risk  
254 factors under analysis include gender, poor education, farming, poor sewage  
255 disposal, visit to water bodies, open dumping of refuse, no BCG vaccination, pet in  
256 the house, poor drinking water source and no preventive measure. Visit to water  
257 bodies is the leading associated risk factor seen in 90% of cases. This agrees with a  
258 similar case-control study done in Ghana [7]. The increased risk for Buruli ulcer  
259 disease may have resulted from direct contact with contaminated water bodies  
260 and/or indirect exposure to riverine environment that harbor *Mycobacterium*  
261 *ulcerans*. The finding also supported previous investigations that noted an elevated  
262 risk for Buruli ulcer disease among Uganda tsetse flies control workers who  
263 frequented swampy areas [8] and among persons who farmed near the Lobo River  
264 in Cote D'ivoire [9]. This finding that regular visit to water bodies such as rivers,  
265 streams and ponds for activities like washing, swimming, fetching, irrigation as  
266 highest risk for Buruli ulcer concurred with study done in Suhum-Kraboia Coalter and  
267 Akuapem South Districts of the Eastern Region of Ghana where presence of wet  
268 land, insect bites in water/mud, washing in the Densu river and house walls built with  
269 mud were identified as risk factors for Buruli ulcer disease [10].

270 The finding of males preponderance of Buruli ulcer disease and lesions on  
271 extremities agrees with findings of Pratima L et al in Ghana [7] but Ernest Kenu et al  
272 found almost equal preponderance between males 49.6% and females 50.4% [10].  
273 The over representation of males suggests that some undiscovered common  
274 behaviour increases their risk for Buruli ulcer disease. The adventurousness of  
275 males and engagement in outdoor activities may account in part for this increased  
276 prevalence in males.

277 Farming was found not to be significantly associated with Buruli ulcer in this study.  
278 This is a surprise finding when considered side by side with many studies which  
279 found Buruli ulcer more commonly among the rural farmers. This may be attributed  
280 to the fact that most people engaged in one form of farming activities or the other  
281 without regarding themselves as farmers. In most rural African towns, it is a common  
282 practice for young children to assist their parents in farming after school hours and/or  
283 on weekend without being labeled as farmers but students.

284 Poor water sources like rivers, streams and ponds were found to be significant risk  
285 factors of Buruli ulcer. This agrees with a study in Ghana assessed water related risk  
286 factors for Buruli ulcer [11]. This finding may not necessarily be related to use of  
287 water from rivers and streams for drinking, cooking, bathing and washing purposes  
288 but likely to be associated with activities such as swimming and playing in the rivers  
289 when fetching water. Persons who fetch boreholes and pipe-borne water are not  
290 likely to engage in these risk activities.

291 The study also found that BCG vaccination significantly reduces the risk for Buruli  
292 ulcer. Studies in Democratic Republic of Congo, Ghana and Togo reported similar  
293 findings in Ghana but contradictory results in DR Congo and Togo [12]. These  
294 discrepancies are explained by three main factors: the BCG strain used for  
295 vaccination, the population vaccinated and Mycobacterium disease. Although BCG  
296 vaccination appears to be effective against leprosy and may protect against Buruli  
297 ulcer disease within 1 year after vaccination, [13] the hypothesis that BCG  
298 vaccination provides lasting protection against Buruli ulcer disease is a matter for  
299 further research.

300 Keeping pets (domestic animals) in the house was found to be significantly  
301 associated with Buruli ulcer disease. There is no tangible explanation for this finding.  
302 It may be a co-incidental finding especially when it is known that Buruli ulcer is  
303 common among people with low socio-economic status. Persons of low socio-  
304 economic status are more likely to keep and live with domestic animals in their  
305 homes.

306 The study also found that non-practice of preventive measures significantly  
307 increases the risk for Mycobacterium Ulceran infections [14]. Bacteria contamination  
308 of skin surfaces may facilitate Mycobacterium ulcerans infection. The regular bathing  
309 may remove bacteria deposited on the skin. This study also corroborates the  
310 observation that wearing protective clothing is associated with decreased Buruli ulcer  
311 disease risk [28] and furnishes important clues about Buruli ulcer disease  
312 transmission. Taken together with the predominance of Limbs lesions suggest that  
313 exposed skin facilitates infection. This study also confirms, however, finding of other  
314 studies that farming with long sleeves and long pants protect against Buruli ulcer [9,  
315 15]. Long clothes may protect from small injuries or insect bites as possible means of

316 entry for *Mycobacterium ulcerans*. The study also found that footwear use was  
317 significantly associated with lower odds of Buruli ulcer and it agrees with findings of  
318 Sara Tomezy K. et al [16]. These findings are consistent with the mode of  
319 transmission of Buruli ulcer which includes presence of skin cut or abrasions and  
320 contact with water, soil or mud during work or recreational activities contaminated  
321 with human and animal excreta.

322 Poor education was found among Buruli ulcer patients, seen in 80% of cases. The  
323 finding is not surprising as Buruli ulcer is a disease associated with poverty, low  
324 socio-economic status and ignorance. A well educated person is more likely to have  
325 a high socio-economic status with high living standard which involves better source  
326 of drinking water, and good personal hygiene.

327 The study found a low level of awareness of Buruli ulcer disease and its risk factors  
328 among cases and control subjects. This agrees with similar study done in Ga West  
329 District of Ghana [17]. However, this was not in keeping with a similar study done in  
330 Southwest Cameroun which found that 84.4% of the studied population knew Buruli  
331 ulcer [6], but lack the knowledge of its aetiology and risk factors. The misconception  
332 found in this study attributing Buruli ulcer to witchcraft was similar to the above  
333 mentioned works and other studies of some African countries. Among the subjects  
334 who knew the cause and risk factors of Buruli ulcer, stratification with respect to level  
335 of education revealed a significant difference in their understanding of the cause of  
336 Buruli ulcer in the favour of well-educated people.

337 Some Buruli ulcer patients categorically stated that their problem was not natural but  
338 rather, the handiwork of evil men. One of the female patients narrated her ordeal in  
339 the hands of a witch doctor who falsely accused her of killing her own mother, and  
340 subsequently extracting a forced confession from her in the mistaken belief that  
341 those were vital steps for cleansing and cure.

342 No community preventive measure/s were identified during the course of this  
343 research and personal hygiene practices found were mostly a part of general  
344 cleanliness and precautions not necessarily against Buruli ulcer.

345 Treatment seeking behaviour could be related to the perception of the cause of the  
346 disease. Majority believed that disease was caused by witchcraft and should be  
347 addressed by traditional healers or witchdoctors. All the patients interviewed have

348 sought treatment from various traditional and herbal practitioners. Majority have  
349 visited various spiritual houses with no visible improvement or cure. This study found  
350 that ignorance and misconceptions were the major causes of delay in seeking  
351 appropriate treatment which led to severe form of the disease and complications,  
352 further supporting the need for intensive health education.

353 In conclusion this study has shown that a significant relationship exists between  
354 Buruli ulcer and the following risk factors: regular visit to water bodies, poor  
355 education, no BCG vaccination and poor drinking water source. Exposure and/or  
356 contamination of skin may facilitate transmission, as suggested by the predominance  
357 of limb lesions and the protective effects of bathing and protective clothing. This  
358 study also clearly demonstrated that there is a wide gap in the public awareness of  
359 Buruli ulcer disease. It also revealed misconceptions about Buruli ulcer aetiology and  
360 mode of transmission which greatly influenced the treatment seeking behaviour.  
361 Proper community education and mobilization are urgently needed to correct the  
362 misconception and improve outcome of Buruli ulcer disease.

363

#### 364 **CONFLICT OF INTEREST**

365 The author(s) declare that there is no conflict of interest regarding the publication of  
366 this paper

367

#### 368 **AUTHOR'S CONTRIBUTIONS**

369 POUA designed the study and supervised the protocol and field study, JCI collected  
370 the data, HNC entered, analyzed and interpreted the data. NBE participated in the  
371 write up for publication

372

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441 **TABLES**

442

443 Table 1: Age and sex distribution of the subjects

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Age group (years)	Case			Control		
	Male	Female	Total	Male	Female	Total
<16	7	1	8	8	8	16
16-25	10	2	12	15	14	29
26-35	5	0	5	13	8	22
>35	6	9	15	5	8	13
Total	28	12	40	41	39	80
Mean age			16.40			16.21
SD			2.10			2.80
Z-test	0.447					
p-value	P>0.05					

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462 Table 2: Distribution of Subjects by Other Demographic Characteristics

Variable	Cases	Control	Total	(X <sup>2</sup> test) p-value (with BfC)*
<b>Education of respondents</b>				
No formal education	12	3		
Primary education	20	32		
Secondary education	8	21		P=0.001**
Tertiary education	0	24		
Total	40	80	120	
<b>Occupation of respondents</b>				
Student	18	35		
Farming	14	16		P=0.85
Civil servant/Teacher	6	19		
Trader/Driver	2	10		
Total	40	80	120	
<b>Comorbidities</b>				
Hypertension	6	9		
Tuberculosis/Diabetes	3	3		P=0.48
None	31	68		
Total	40	80	120	
<b>Social history</b>				
Smokes	1	3		
Drinks alcohol	15	27		P= 0.35
None significant	24	50		
Total	40	80	120	

463

464 \*BfC – Bonferoni Correction; \*\*statistically significant

465 Table 3: Distribution of respondents according to other socio-demographic  
466 characteristics

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				( $X^2$ test)
Variables	Cases	Control	Total	p-value (with BfC)*
<b>Sewage disposal method</b>				
Pit toilet	24	40		
Water closet	4	24		P=0.7
Bucket	1	4		
Bush/open	11	12		
Total	40	80	120	
<b>Sources of drinking water</b>				
Stream/River/water body	31	12		
Pipe borne water	2	39		P=0.01**
Well	3	10		
Borehole	1	9		
Sachet/bottled water	3	10		
Total	40	80	120	
<b>Type of living house</b>				
Public	19	14		
Private	21	66		P=0.001**
Total	40	80	120	
<b>Refuse disposal method</b>				
Open	28	53		
Close	12	27		P=0.1
Total	40	80	120	
<b>Pet in the house</b>				
Yes	18	15		
No	22	65		P=0.015**
Total	40	80	120	
<b>BCG vaccination</b>				

Yes	16	52	
No	24	28	P=0.03**
Total	40	80	120
<b>BCG Scar present</b>			
Yes	14	51	
No	2	1	P=0.6
Total	16	52	68
<b>Visit to water body</b>			
Yes	36	10	
No	2	70	P=0.0001**
Total	40	80	120

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470 \*BfC – Bonferoni Correction; \*\*Statistically significant

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490 Table 4: Awareness, characteristics and management of the ulcer among the cases

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Variables	Cases	% of total	Control	% of total
<b>Heard about Buruli ulcer</b>				
Yes	2	5%	1	1.25%
No	38	95%	79	98.75%
Total	40		80	
<b>Knowledge of risk factors</b>				
No idea	38	95%	78	97.50%
Acquired from the ground	2	5%	2	2.50%
Total	40		80	
<b>Site of lesion</b>				
Head	0	0%	NA	
Upper limb	12	30%	NA	
Trunk	0	0%	NA	
Lower limb	28	70%	NA	
Total	40			
<b>Duration of lesion</b>				
≤8 months	2	5%	NA	
>8 months	38	95%	NA	
Total	40			
<b>Preventive measures adopted**</b>				
None	20	50%	11	13.75%
Bathes after work	6	15%	27	33.75%
Bathes daily	5	12.5%	22	27.50%
Wears foot wear always	5	15%	15	18.75%
Wear protective clothing	4	10%	10	12.50%
Uses ITN	11	27%	32	40.00%
<b>Treatment of ulcer before coming to hospital*</b>				

Herbal concoctions	38	95%	NA	
Spiritual homes	30	75%	NA	
Wound dressing in local patent medicine store	39	97%	NA	

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493 \*\*Multiple responses for those who practice personal protection

494 \*Multiple responses

495 NA – Not applicable

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EARLY VIEW

520 Table 5: Relationship between Buruli ulcer and the common risk factors

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Buruli Ulcer Risk factors	Case	Control	Total OR	X <sup>2</sup>	p-value
Gender (male)	28(70.0%)	41(51.3%)	69 2.22	3.84	=0.05*
Poor Education	32(80.0%)	35(43.8%)	67 5.13	14.27	=0.0003*
Farming	14(35.0%)	16(20.0%)	30 2.15	3.20	=0.07
Poor sewage disposal	12(30.0%)	16(20.0%)	28 1.71	1.49	=0.15
Visit to water bodies	36(90.0%)	10(12.5%)	46. 63	67.78.	=0.00000*
Open dumping of refuse	28(70.0%)	53(66.3%)	81 1.19	0.17	=0.83.
No BCG vaccination	24(60.0%)	28(35.0%)	52. 2.79	6.79.	=0.01*
Pet in the house	18(45.0%)	15(18.8%)	33. 3.55	9.25.	=0.002*
Poor drinking water source	31(77.5%)	12(15.0%)	43. 19.52	45.32.	=0.00000*
No preventive measure	20(50.0%)	11(13.8%)	31. 6.27	18.30.	=0.00005*

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523 \*Statistically significant