Full Length Research Paper

Environmental and behavioural factors associated with Mycobacterium ulcerans infection in the district of Lalo in Benin: A case-control study

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The mode of transmission of Buruli ulcer (BU) is not yet well understood. This study aimed to identify risk factors for BU in the district of Lalo in Benin. This is a case-control study taking into account all cases of BU detected from 2013 to 2018 and treated at the Buruli’s ulcer Screening and Treatment Center (CDTUB) of Lalo. For each case, two controls were matched according to age and sex. Conditional logistic regression was used to compare risk factors in cases and controls. A total of 59 BU cases and 118 controls were enrolled. After adjustment for potential confounding factors, illiteracy (adjusted odd ratio [aOR] = 18.19; 95% confidence interval [CI] = 1.50-219.45), unawareness of BU risk factors (aOR = 48.21; 95% CI = 4.67-497.15), bathing in rivers or ponds (aOR = 23.66; 95% CI = 2.82-198.52), field activities in mud (aOR = 60.58; 95% CI = 7.44 – 493.09) and not wearing protective clothing during field activities (aOR = 42.78; 95% CI = 2.05 – 892.33) were associated with BU. It is necessary to mitigate risk factors by implementing actions in the field of water, sanitation and hygiene (WASH) and by insisting on the wearing of protective equipment during agricultural activities and/or contact with surface water.

Key words: Buruli ulcer, Mycobacterium ulcerans, risk factors, case-control study, Lalo.

INTRODUCTION

Mycobacterium ulcerans (Mu) is the pathogen responsible for Buruli ulcer (BU), a neglected tropical disease (Gangbo et al., 2011). It is the third most common mycobacterial disease (after leprosy and tuberculosis) that affects immunocompetent people worldwide (Hotz and Kamath, 2009). It affects people of all ages – mostly children (Stoffel et al., 2005; Kanga et al., 2006) – and occurs in various forms. The most serious forms are extensive skin ulcerations, sometimes leading to disabling sequelae (Adu and Ampadu, 2015). This disease has
been reported in 33 countries worldwide, with the majority of cases in the tropical zone of West and Central Africa (Hotez et al., 2007; World Health Organization, 2015). Studies have also identified the disease in Australia, China and Japan (Huang and Johnson, 2014). In Benin, BU was first diagnosed in 1977 by Sister Aguiar at the Saint Camille Hospital of Dogbo in the Couffo Department. Currently, it is endemic in eight of the twelve departments in Benin: the Atlantic, the Collines, the Couffo, the Littoral, the Mono, the Ouémé, the Plateau and the Zou (Johnson et al., 2005). These departments are located in the south of the country. From 1989 to 2019, more than 13,000 new cases were detected (PNLLUB, 2019). The emergence of the disease over the past 50 years, as well as its geographic distribution, has been largely associated with aquatic ecosystems, particularly in areas with slow and stagnant water flow (Wagner et al., 2008; Marion et al., 2011). 

The epidemiology of BU is not yet well known (Portaels et al., 2001; WHO, 2017), but studies in several endemic regions have associated its occurrence with environmental disturbances resulting from human activities such as dam construction, mining, construction of artificial lakes and extension of swamps for rice and fish farming (Brou et al., 2008; Wagner et al., 2008b; Landier et al., 2014; Aboagye et al., 2017).

The exact mode of transmission of Mu to humans is unclear; however, several studies have shown that the pathogen is found in aquatic ecosystems (Wagner et al., 2008a; Marion et al., 2011). Since its transmission is not clearly understood in endemic communities, potential environmental and behavioural risk factors identification can help define preventive measures to reduce the number of cases. Potential risk factors appear to be geographically specific. In Ghana, for example, practicing agriculture in swampy areas, land cultivation in short outfits, insect bites, applying leaves to wounds plus wading in rivers, have been identified in studies as risk factors (Raghunathan et al., 2005a; Aboagye et al., 2017). In Tissalé, Ivory Coast, it was identified that regular contact with unprotected surface water and lack of protective equipment during agricultural activities were the main factors associated with the risk of contracting BU (N’krumah et al., 2016). In Togo, factors such as age, receiving insect bites near a river in older children, and bathing with water from open-pit drilling remain potential risk factors that increased Mu infection (Maman et al., 2018b). In Australia, the risk of BU was associated with mosquito bites on the lower legs or arms (Quek et al., 2007). These studies show that the risk factors associated with BU tend to vary from one locality to another. This could probably be due to geographical, environmental and behavioural differences. In Benin, few studies have specifically addressed BU risk factors. Thus, the present case-control study aims to elucidate the potential environmental and behavioural risk factors in the district of Lalo, known as one of the most endemic districts for BU in Benin.

MATERIALS AND METHODS

Study framework

The study is conducted in southern Benin, in the district of Lalo located in the department of Couffo, in the south of the country. Notably, 746 of the 4,817 cases of BU screened from January, 1st 2008 to December, 31st 2018 came from this district (PNLLUB, 2018). Partitioned into 11 subdistricts and 61 villages, Lalo’s district is one of the six administrative subdivisions of the department of Couffo and covers an area of 432 km², or 0.8% of the total area of Benin (Biaou, 2006). Its population is estimated at approximately 138,451 based on projections from 2018 (INSAE, 2013) (Figure 1).

Lalo has a Guinean Sudanese-type climate, characterized by small temperature differences of approximately 27°C (Biaou, 2006). This type of climate allows it to have a succession of four seasons per year (an alternation of two dry and two rainy seasons). The annual average rainfall ranges from 900 to 1,100 mm, and the relative humidity can reach 85% (Biaou, 2006). In recent decades, rains have become increasingly random, and drought sequences are observed in the middle of the rainy season. The months of December and January suffer the increasing depression of the harmattan, whose breath dries and promotes a high thermal amplitude during the day. Like most municipalities in the Couffo department, it is located on the Aplahoué plateau. The morphological and topographical study of this relief shows that it is a clay plateau region with an average altitude of 80 m ending in a clay depression: Tchi depression (Biaou, 2006). These climatic conditions and soil types that characterize the district favour dense vegetation, the presence of rivers, lakes, slow-flowing rivers, ponds and swampy areas that are favourite areas of Mu, the pathogen responsible for the BU. The economic activities of the people in this commune are mainly agriculture and trade affairs.

Study design and population

This is a case-control study taking into account all cases of BU detected from January, 1st 2013 to December, 31st 2018 in the district of Lalo and handled at the Buruli Ulcer Screening and Treatment Center (CDTUB) of Lalo and confirmed by Polymerase Chain Reaction (PCR). In each of the identified cases, two controls are matched.

Participants

A case has been defined as any person living in the district of Lalo and showing off clinical signs of the disease as defined by the WHO...
Figure 1. Administrative map of Lalo district.

Data collection

The patients included in the study were recruited retrospectively from Lalo’s Buruli Ulcer Screening and Treatment Center database. They have been actively identified in the community. Recruitment was voluntary; trained people went to the homes of the selected cases to obtain their participation consent. After explanation of the study’s purpose to the participants and of their consent, two matched controls were randomly selected from the same environment as the case (neighbourhood, village or hamlet). If one of the participants was not available, a second pass was scheduled. A standardized questionnaire was administered to all selected participants. For those who could not answer in French, the interview was conducted in the local language. For children under the age of ten, most of the responses, particularly those related to occupation and lifestyle in the face of illness, were given by the
parents.

Variables

The variables involved in this study relate to demography, environment and environmental practices. We also studied outdoor behaviour and habits, including clothing worn, occupations and activities associated with the aquatic environment. Case were asked to limit their responses to the year prior to the onset of their first signs of the disease; the same condition was also explained to those of the control group. Contacts who were fluent in the participants’ native language administered the questionnaires with the help of community volunteers.

Data processing and statistical analysis

Data collected were captured twice using Microsoft Excel 2010 software and analysed using Epi Info® 7 and Stata 14 software. BU was the dependent variable, while sociodemographic factors, knowledge, attitude and practices of participants, lifestyles and workplaces in the environment were independent variables. All variables were described by proportions. A univariate analysis was performed to describe the association between BU and the independent variables. The significance threshold was 5%. Variables with a P-value less than or equal to 0.2 and highlighted by other studies as associated variables were selected for multivariate analyses. Conditional logistic regression was used with a progressive top-down elimination process to identify variables significantly associated with BU to control for possible confounding factors in the model. P-values of 0.05 were considered statistically significant.

Ethical aspects

The present study was authorized by the health authorities of the National Buruli Ulcer and Leprosy Control Program (PNLULUB) of Benin and by those of the district of Lalo. The participants gave their verbal consent. The data were treated with strict respect for confidentiality and anonymity.

RESULTS

Sociodemographic characteristics of participants

A total of 59 BU cases and 118 controls were enrolled in this study (Table 1). Of the 59 BU cases, 27(45.76%) were male, so the sex/ratio (male/female) was 0.8. The median age of the cases was 12 years (Q1 = 6; Q3 = 28), ranging from 1 to 60 years. The majority of them were under the age of 15 (35 cases or 59.32%) and were single (42 cases or 72.2%). Their main activity was agriculture (52 cases or 88.14%). Of the 59 BU cases surveyed, 33 (55.93%) had not received formal education. Of the 118 controls, 54 (45.76%) were male, so the sex/ratio (male/female) was 0.8. The median age was 13 years (Q1 = 7; Q3 = 26), ranging from 1 to 60 years. More than half of them were under the age of 15 (69 or 58.47%), and 81 were single (68.64%). Their main activity was agriculture (96 or 81.36%). The data showed that 54 (45.76%) had not received formal schooling.

Clinical characteristics of cases

All cases from the study were confirmed by PCR. Based on Table 2, the predominant clinical form is the ulcerated form (43 cases or 72.88%). Most lesions were present in the lower limbs (30 cases or 50.85%). Serious lesions (category 3) accounted for 33.9%.

Univariate analysis of risk factors for contracting Buruli ulcer in the district of Lalo in Benin

Sociodemographic factors linked with BU

Illiteracy was significantly associated with a higher risk of developing BU (OR=5.35-95%; CI=1.77-16.15) (Table 3). A lack of knowledge of the causes (OR = 2.80; 95% CI = 1.34-5.88) and risk factors for the disease (OR = 5.56; 95% CI = 2.08 - 14.90) is positively associated with its onset.

Environmental and behavioural factors linked to BU onset

Field activities in the mud significantly increased the risk of developing BU (OR = 5.67; 95% CI = 2.95-10.87). Individuals who used surface water sources (OR = 3.89; 95% CI = 1.46-10.34) and baths in rivers or ponds (OR = 4.71; 95% CI = 2.26-9.80) were much more likely to contract BU (Table 3). However, wearing footwear during field activities (OR = 0.15; 95% CI = 0.04-0.54) and wearing protective clothing during cultivation (OR = 5.10; 95% CI = 1.48 -17.56) significantly reduced this risk.

Multivariate analysis of associated risk factors for contracting Buruli ulcer in the district of Lalo in Benin

After adjusting for potential confounding factors, we identified that illiteracy (aOR = 18.19; 95% CI = 1.50-219.45), lack of awareness of BU risk factors (aOR = 48.21; 95% CI = 4.67-497.15), swimming in rivers or ponds (aOR = 23.66; 95% CI = 2.82-198.52), field activities in mud (aOR = 60.58; 95% CI = 7.44 – 493.09) and not wearing protective clothing during field activities (aOR = 42.78; 95% CI = 2.05 – 892.33) were associated with the occurrence of BU (Table 4).

DISCUSSION

Our results showed that poor knowledge of the causes
and risk factors for BU increases the risk of developing the disease. This situation could be explained by the low level of education of the populations of the district, which is a rural area (Biaou, 2006). Indeed, in the framework of our study, uneducated subjects have a significantly higher risk of having BU than those whose educational level is higher or equal to primary school. These results are similar to those of a study conducted in Cameroon (Pouillot et al., 2007). According to these authors, the low level of education (lower to secondary) of the subjects involved in their study was observed as a significant risk factor for BU. Therefore, improving education for populations in endemic areas would reduce the risk of BU. Studies have indeed shown that education improves health because it increases the effectiveness of action, strengthening the sense of personal control that encourages and enables a healthy lifestyle (Ross and Wu, 1995; Hahn and Truman, 2015).

Our analyses did not reveal a significant association between the proximity of water bodies to the subjects'
Table 2. Clinical characteristics of BU cases.

| Variables           | Effective (n) | Frequency (%) |
|---------------------|---------------|---------------|
| **Clinical form**   |               |               |
| Nodule              | 1             | 1.69          |
| Plaque              | 13            | 22.03         |
| Ulcer               | 43            | 72.88         |
| Osteomyelitis       | 2             | 3.39          |
| **Localization**    |               |               |
| UP                  | 23            | 38.98         |
| LL                  | 30            | 50.85         |
| Others              | 6             | 10.17         |
| **Category**        |               |               |
| Category 1          | 6             | 10.17         |
| Category 2          | 33            | 55.93         |
| Category 3          | 20            | 33.9          |

*Localization: UP = Upper limbs; LL = Lower limbs; Others = Abdomen, Thorax, Back.

Table 3. Univariate analysis of chosen variables linked to Buruli ulcer in Lalo’s district, Benin (2013 – 2018).

| Characteristics                  | Cases n (%) | Controls n (%) | OR (IC 95%)      | P-value |
|----------------------------------|-------------|----------------|------------------|---------|
| **Education**                    |             |                |                  |         |
| Uneducated                       | 33 (55.93)  | 43 (36.44)     | 5.35 (1.77 - 16.15) | 0.003   |
| Educated (Elementary and high school) | 26 (44.07)  | 75 (63.56)     | 1                |         |
| **Ethnic**                       |             |                |                  |         |
| Fon and relatives                | 29 (49.15)  | 60 (50.95)     | 1                |         |
| Adja and relatives               | 30 (50.85)  | 58 (49.15)     | 1.34 (0.36 - 4.92) | 0.656   |
| **Occupation**                   |             |                |                  |         |
| No occupation                    | 3 (5.08)    | 4 (3.39)       | 5.05 (0.62 - 40.72) | 0.128   |
| Farmers                          | 52 (88.14)  | 96 (81.36)     | 3.37 (0.88 - 12.79) | 0.074   |
| Others                           | 4 (6.78)    | 18 (15.25)     | 1                |         |
| **House wall hardware**          |             |                |                  |         |
| Branches or slabs wall           | 10 (16.95)  | 22 (18.64)     | 1.08 (0.22 - 5.31) | 0.922   |
| Clay wall                        | 43 (72.88)  | 77 (65.26)     | 1.77 (0.66 - 4.73) | 0.253   |
| Brick or metal wall              | 6 (10.17)   | 19 (16.10)     | 1                |         |
| **Roof-covering**                |             |                |                  |         |
| Straw roof                       | 11 (18.64)  | 22 (18.64)     | 1 (0.42 - 2.33)   | 1       |
| Slab or metal                    | 48 (81.36)  | 96 (81.36)     | 1                |         |
| **Proximity swamps and dwellings**|           |                |                  |         |
| Yes                              | 34 (57.63)  | 63 (53.39)     | 1.54 (0.56 - 4.17) | 0.394   |
| No                               | 25 (43.37)  | 55 (46.61)     | 1                |         |
| **Proximity rivers/watercourse and dwellings** | | | | |
| Yes                              | 22 (37.29)  | 49 (41.53)     | 0.68 (0.26 - 1.76) | 0.431   |
Table 3. Contd.

|                          | No                      | Yes                      | p-value |
|--------------------------|-------------------------|--------------------------|---------|
| Pile of junk near houses |                         |                          |         |
| Yes                      | 37 (62.71)              | 49 (83.05)               | 0.66 (0.25 - 1.73) | 0.407   |
| No                       | 69 (58.47)              | 103 (87.29)              | 1       |
| Field activities in mud  |                         |                          |         |
| Yes                      | 1 (23.73)               | 45 (76.27)               | 5.67 (2.95 - 10.87) | < 0.001 |
| No                       | 10 (16.95)              | 29 (24.58)               | 1       |
| Footwear while working   |                         |                          |         |
| Yes                      | 6 (11.54)               | 32 (33.33)               | 0.15 (0.04 - 0.54) | 0.004   |
| No                       | 46 (88.46)              | 64 (66.67)               | 1       |
| Wearing protective outfits during field activities | | | |
| Yes                      | 3 (5.08)                | 26 (22.03)               | 1       |
| No                       | 56 (94.92)              | 92 (77.97)               | 5.10 (1.48 - 17.56) | 0.002   |
| Water supply             |                         |                          |         |
| Surface water            | 14 (23.73)              | 10 (8.47)                | 3.89 (1.46 - 10.34) | 0.006   |
| Drillings / wells        | 45 (76.27)              | 108 (91.53)              | 1       |
| Swimming into rivers/ponds |                       |                          |         |
| Yes                      | 30 (50.85)              | 20 (19.95)               | 4.71 (2.26 - 9.80) | <0.001  |
| No                       | 29 (49.15)              | 98 (83.05)               | 1       |
| Understanding of causes  |                         |                          |         |
| Low                      | 32 (54.24)              | 40 (30.90)               | 2.80 (1.34 - 5.88) | 0.006   |
| Good                     | 27 (45.76)              | 78 (66.10)               | 1       |
| Risk factors understanding |                       |                          |         |
| Low                      | 49 (83.05)              | 68 (57.63)               | 5.56 (2.08 - 14.90) | 0.001   |
| Good                     | 10 (16.95)              | 50 (42.37)               | 1       |

homes and the risk of contracting BU. The pathogen *Mu*, which is responsible for BU, is found in the aquatic environment of endemic areas, as demonstrated by several studies (Wagner et al., 2008a; Marion et al., 2011; Maman et al., 2018a). Therefore, the disease could only occur through direct and frequent contact of the populations with this contaminated environment. Indeed, swimming in rivers and ponds has been identified as being significantly associated with the risk of contracting BU in the district of Lalo. Similar results were also found in Togo (Maman et al., 2018b), Ghana (Raghunathan et al., 2005b; Kenu et al., 2014), the Ivory Coast (N’krumah et al., 2016) and Cameroon (Pouillot et al., 2007). These daily life behaviours of the subjects involve direct contact with the aquatic environment that houses *Mu*, thus increasing contamination risks.

Univariate analysis revealed that being a grower is not significantly associated with the risk of BU. However, farming in mud significantly increases the risk of contracting BU. Our results are consistent with other epidemiological studies conducted in BU endemic areas (Marston et al., 1995; Kenu et al., 2014; Wu et al., 2015; Aboagye et al., 2017). The cases of BU involved in our study are mostly growers (88%); among them, many conduct their farming activities in the mud as well as the controls. Agricultural activities in wetlands or even swampy areas put these cases in frequent contact with wet or muddy soils. Studies have been able to identify *Mu* strains in soil and decompose organic matter under water, and they can sometimes live for several months
Table 4. Multivariate model of risk factors for Buruli ulcer in the district of Lalo in Benin (2013 – 2018).

| Variables                        | Adjusted OR (IC à 95%) | P-value |
|----------------------------------|------------------------|---------|
| Education                        |                        |         |
| Educated (Elementary / High school) | 1                      |         |
| Uneducated                        | 18.19 (1.50 - 219.45)  | 0.022   |
| Wearing protective outfits during field activities |                        |         |
| Yes                              | 1                      |         |
| No                               | 42.78 (2.05 - 892.33)  | 0.015   |
| Field activities in mud          |                        |         |
| Yes                              | 60.58 (7.44 - 493.09)  | < 0.001 |
| No                               | 1                      |         |
| Swimming into rivers/ponds       |                        |         |
| Yes                              | 23.66 (2.82 - 198.52)  | 0.004   |
| No                               | 1                      |         |
| Risk factors understanding       |                        |         |
| Bas                              | 48.21 (4.67 - 497.15)  | 0.001   |
| Good                             | 1                      |         |

(Bratschi et al., 2014; Aboagye et al., 2016; Tian et al., 2016a, b; Röltgen et al., 2017). This may lead us to believe that BU cases may have become infected with muddy soils through micro trauma or open wounds on their skin (Yotsu et al., 2015). These data clearly confirm that agriculture in wetlands or swampy areas is a potential risk factor for BU. In addition, we found that wearing protective clothing during field activities guards them against BU. These results are consistent with several studies (N’krumah et al., 2016; Aboagye et al., 2017; Maman et al., 2018). In fact, most people having BU and involved in farming do not wear protective outfits. This may be due to the frequent warm weather conditions in most African countries that encourage farmers to wear less protective clothing during farming to work at ease (Aboagye et al., 2017). Data from the literature have shown that Buruli ulcer is much more prevalent in youth younger than 15 years of age (World Health Organization, 2012; Yotsu et al., 2015); our results were similar. Nevertheless, some study results differ from our findings. Indeed, the results of a study conducted in Nigeria over 2014 and 2015 showed a predominance of the disease in subjects 15 years old (Meka et al., 2016). Note that in Australia, some studies have shown a predominance of the disease in adult subjects (Yerramilli et al., 2017; Walker et al., 2019). This discrepancy between our study and that of Nigeria could be explained by differences between the study periods. In our context, the study was broader (12 years), which allowed us to have much more data for analysis, unlike that of Nigeria (May 2014 to September 2015 or approximately 16 months). Dissimilarities between our study and those conducted in Australia could be explained by two factors. First, we can hypothesize that children in Africa are much more exposed to the aquatic environment than those in Australia through recreational activities such as walking or bathing in streams or bodies of water. Second, it has been postulated that African strains of Mu are more virulent than strains endemic to Australia (Mve-Obiang et al., 2003). It is important to note, however, that many other factors, such as genetic factors, socioeconomic context, cultural views on disease and the availability of health care facilities, play an important role in determining the severity of Buruli ulcer.

Case-control studies have several limitations, including the risk of recall bias. Twenty-five of the patients were under the age of 10. For this category of participants, both cases and controls, the answers to the questions were given by the parents/tutors and may have been biased. Our study may also suffer from selection bias, as we only recruited cases from the district of Lalo and took over some from the CDTUB of Lalo. Training and awareness sessions were conducted, and participants in our study may have already received basic information about the disease. This is likely to introduce recall bias. We tried to minimize this bias in the current study by using a well-designed (culturally and contextually sensitive) and pretested questionnaire. Investigators
were also trained to ensure that appropriate responses were obtained from respondents to minimize any form of bias or confounding effects in the results.

CONCLUSION

Our study has shown that contact with unprotected water (surface water, swamps and wetlands) in daily activities such as agriculture and swimming is the main factor associated with Buruli ulcer. This factor would be favoured by the low level of education of the populations and the unawareness or lack of knowledge of the disease’s risk factors. It is important to mitigate risk factors in endemic communities by implementing actions in the field of water, sanitation and hygiene (WASH) and emphasizing the use of protective equipment and clothing during rural activities and/or contact with surface water.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Aboagye SY, Asare P, Otchere ID, Koka E, Mensah GE, Yirenya-Tawiah D, Yeoob-Manu D (2017). Environmental and Behavioral Drivers of Buruli Ulcer Disease in Selected Communities along the Denzu River Basin of Ghana: A Case-Control Study. The American Journal of Tropical Medicine and Hygiene 96(5):1076-1083. https://doi.org/10.4269/ajtmh.16-0749

Aboagye SY, Danso E, Ampah KA, Nakobu Z, Asare P, Otchere ID, Rößgen K, Yirenya-Tawiah D, Yeoob-Manu D (2016). Isolation of Nontuberculous Mycobacteria from the Environment of Ghanaian Communities Where Buruli Ulcer Is Endemic. Applied and Environmental Microbiology 82(14):4320-4329. https://doi.org/10.1128/AEM.01002-16

Adu EJK, Ampadu E (2015). Mycobacterium ulcerans disease in the middle belt of Ghana: An eight-year review from six endemic districts. International Journal of Mycobacteriology 4(2):138-142. https://doi.org/10.1016/j.ijmyco.2015.03.006

Blaou CF (2006) Monographie de la commune de Lalo. 68p

Bratschi MW, Ruf M-T, Andreoli A, Minyem JC, Kerber S, Wantong FG, Pritchard J, Chakwera V, Beuret C, Wittmer M, Noumen D, Schürch N, Um Book A, Puschke G (2014). Mycobacterium ulcerans persistence at a village water source of Buruli ulcer patients. PLoS Neglected Tropical Diseases 8(3):2756. https://doi.org/10.1371/journal.pntd.0002756

Brou T, Brouthin E, Elguero E, Asse H, Guegan J-F (2008). Landscape Diversity Related to Buruli Ulcer Disease in Côte d’Ivoire. PLoS Neglected Tropical Diseases 2(7):271. https://doi.org/10.1371/journal.pntd.0000271

Gangbo F, Brun LV, Sopoh GE, Johnson RC, Ayadij E, Barogui YT, Houézo JG, Kestens L, Anagoun SY (2011), Aspects épidémiologiques, cliniques et histopathologiques de l’ulcère de Buruli traité pendant quatre ou huit semaines par la streptomycine associée à la rifampicine: à propos de 66 cas. JAMO 51(2):11

Hahn RA, Truman BI (2015). Education Improves Public Health and Promotes Health Equity. International Journal of Health Services: planning, administration, evaluation 45(4):657-678. https://doi.org/10.1177/00207373145585986

Hotez PJ, Kamath A (2009). Neglected Tropical Diseases in Sub-Saharan Africa: Review of Their Prevalence, Distribution, and Disease Burden. PLoS Neglected Tropical Diseases 3(8):412. https://doi.org/10.1371/journal.pntd.0000412

Hotez PJ, Molyneux DH, Fenwick A, Kumsran J, Sachs SE, Sachs JD, Savioi L (2007). Control of neglected tropical diseases. The New England Journal of Medicine 357(10):1018-1027. https://doi.org/10.1056/NEJMra064142

Huang SKL, Johnson PDR (2014). Epidemiology and management of Buruli ulcer. Expert Review of Anti-infective Therapy 12(7):855-865. https://doi.org/10.1586/14787212.2014.910113

INSAE (2013). Quatrième recensement général de la population et de l’habitat : résumé de la synthèse des résultats. Ministère du Développement, de l’Analyse Économique et de la Prospective, Cotonou, Bénin, p. 4, Report No.: RGHP-4

Johnson RC, Sopoh GE, Boko M, Zinsou C, Gbovi J, Makoutode M, Portaels F (2005). Distribution of the infection to Mycobacterium ulcerans (Ulcère de Buruli) dans la commune de Lalo au Bénin. Tropical Medicine and International Health 10(9):863-871

Kanga JM, Kacou ED, Kouamé K, Kassi K, Kaloga M, Yao JK, Dione-Lainé M, Avoaka LE, Yoboué-Yao P, Sangaré A, Ecra JE, Ahogo C, Djédjé MS, Dadiri AJ, Ayé C (2006). [Fighting against Buruli ulcer: the Côte-d’Ivoire experience]. Bulletin de la Société de Pathologie Exotique (1990) 99(1):34-38

Kengne E, Nyarko KM, Schetzfield L, Ganu V, Käsir A, Martey M, Calys-Tagoe BNL, Koram K, Adanu R, Razum O, Afari E, Biinka FN (2014). Risk Factors for Buruli Ulcer in Ghana—A Case Control Study in the Suhum-Krabo-Coaltar and Akuapem South Districts of the Eastern Region. PLoS Neglected Tropical Diseases 11(8):3279. https://doi.org/10.1371/journal.pntd.0003279

Lander J, Gaudent J, Carolan K, Lo Seen D, Guégan J-F, Eyangoh S, Fontanet A, Texier G (2014). Spatio-temporal patterns and Landscape-Associated Risk of Buruli Ulcer in Akonolinga, Cameroon. PLoS Neglected Tropical Diseases 8(9):3123. https://doi.org/10.1371/journal.pntd.0003123

Maman I, Tchacondo T, Kere AB, Beissner M, Badziklou K, Tedihou E, Gangbo F, Brun LV, Sopoh GE, Johnson RC, Ayadij E, Barogui YT, Wiedemann FX, Kanga JM, Kacou ED, Kouamé K, Kaloga M, Yao JK, Dione-Lainé M, Avoaka LE, Yoboué-Yao P, Sangaré A, Ecra JE, Ahogo C, Djédjé MS, Dadiri AJ, Ayé C (2006). [Fighting against Buruli ulcer: the Côte-d’Ivoire experience]. Bulletin de la Société de Pathologie Exotique (1990) 99(1):34-38

Maman I, Tchacondo T, Kere AB, Beissner M, Badziklou K, Wiedemann FX, Karou DS, Bretzel G (2015). Molecular detection of Mycobacterium ulcerans in the environment and its relationship with Buruli ulcer occurrence in Zio and Yoto districts of maritime region in Togo. PLoS Neglected Tropical Diseases 12(5). https://doi.org/10.1371/journal.pntd.0006455

Maman I, Tchacondo T, Kere AB, Piten E, Beissner M, Kobara Y, Kossi K, Badziklou K, Wiedemann FX, Amekuse K, Bretzel G, Karou DS (2018b). Risk factors for Mycobacterium ulcerans infection (Buruli Ulcer) in the maritime region. BMC Infectious Diseases 18. https://doi.org/10.1186/s12879-018-2958-3

Marion E, Lander J, Boisier P, Marsolier L, Fontanet A, Le Gall P, Aubry J, Djenga N, Umboock A, Eyangoh S (2011). Geographic expansion of Buruli Ulcer disease, Cameroon. Emerging Infectious Diseases 17(3):551-552

Marston BJ, Diallo MO, Jr CRH, Diomande I, Saki MZ, Kanga J-M, Partain JT, Lipman HB, BMC SM, Infect RC (1995). Emergence of Buruli Ulcer Disease in the Daloa Region of Cote D’Ivoire. The American Journal of Tropical Medicine and Hygiene 52(3):219-224.
