

# *Mycobacterium ulcerans* infection: control, diagnosis, and treatment

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The skin disease Buruli ulcer, caused by *Mycobacterium ulcerans*, is the third most common mycobacterial disease after tuberculosis and leprosy and mainly affects remote rural African communities. Although the disease is known to be linked to contaminated water, the mode of transmission is not yet understood, which makes it difficult to propose control interventions. The disease is usually detected in its later stages, when it has caused substantial damage and disability. Surgery remains the treatment of choice. Although easy and effective in the early stages of the disease, treatment requires extended excisions and long hospitalisation for the advanced forms of the disease. Currently, no antibiotic treatment has proven effective for all forms of *M ulcerans* infection and research into a new vaccine is urgently needed. While the scientific community works on developing non-invasive and rapid diagnostic tools, the governments of endemic countries should implement active case finding and health education strategies in their affected communities to detect the disease in its early stages. We review the diagnosis, treatment, and control of Buruli ulcer and list priorities for research and development.

## Introduction

Buruli ulcer is primarily an infection of the subcutaneous fat caused by the environmental pathogen *Mycobacterium ulcerans*. Although the disease has a low mortality rate, it has a huge socioeconomic impact on affected populations and is a public-health issue in terms of morbidity, treatment, and functional disabilities.

The clinical lesion of Buruli ulcer usually starts as a painless subcutaneous nodule that secondarily ulcerates, presenting characteristic undermined edges (figure 1). Other pre-ulcerative forms are papules affecting only the skin (observed more often in Australia than other parts of the world), plaques (large firm, painless, and raised lesions), and oedema (severe form of the disease).

*M ulcerans* produces a toxin, mycolactone, which induces necrosis and ulceration by its cytotoxic and immunosuppressive properties. Some patients develop bone (osteomyelitis) and joint lesions through contiguous or haematogenous spread of *M ulcerans*. Later in the natural history of the disease spontaneous healing can occur, although the mechanisms of this spontaneous healing are unclear.<sup>1</sup> Healing takes months and can cause deep scarring, retractions, and deformity. Death is a rare complication. Various recurrence rates after surgical treatment have been reported, from 6·1% within 7 years of follow-up<sup>2</sup> to 16% after a follow-up period of 1 year.<sup>3</sup>

Since 1980, there has been a large increase in the detection rate of the disease. Buruli ulcer is now the third most common mycobacterial infection in human beings after tuberculosis and leprosy, and is the most poorly understood of the three diseases. The disease often occurs in localised and remote areas where populations have limited access to medical care. The median age of Buruli ulcer patients is about 15 years; the distribution of age-specific detection rates peaks at 10–14 years and 75–79 years.<sup>4</sup> Buruli ulcer has been reported or suspected in 30 tropical countries worldwide,

including Australia, but west Africa is the most affected region (figure 2).<sup>6</sup> Cases have also been reported in non-tropical areas.<sup>6</sup> The number of cases reported in some countries of west Africa is substantial: 5700 between 1989 and 2003 in Benin, with a Buruli ulcer detection rate of 21·5 per 100 000 in 1999 in the Zou region;<sup>7</sup> 17 000 between 1978 and 2003 in Côte d'Ivoire, where prevalence reaches 16% in some villages;<sup>8</sup> in Ghana, a national survey done in 1999 detected 5619 cases (overall crude national prevalence of 20·7 per 100 000 but 158·8 per 100 000 in the most affected districts), of which



Figure 1: A typical Buruli ulcer



Figure 2: Distribution of Buruli ulcer worldwide

Red dots show the approximate location of disease within affected countries. Adapted from reference 5.

almost 50% were at the ulcerative stage.<sup>9</sup> However, most of these data come from passive case detection and are probably under-estimates of the extent of the problem worldwide.<sup>10</sup>

Part of the explanation for the emerging character of the disease could be an increase in public and scientific awareness. Buruli ulcer has long gone largely unnoticed, possibly because it occurs in remote rural communities that are poorly covered by national health surveillance systems. Additionally, many of the affected populations do not seek health services because of financial constraints, beliefs that the treatment does not work, fear of surgery and anaesthesia, or superstition and stigma.<sup>11,12</sup> However, some researchers<sup>7,13</sup> have concluded that the emergence of the disease is not only detection bias but also an increase in the exposure of affected populations. Outbreaks appear to be related to environmental changes (deforestation, agriculture, and hydraulic installations) involving surface water.<sup>14</sup>

Areas affected by Buruli ulcer disease are located near stagnant or slow-flowing water areas. Despite epidemiological<sup>15</sup> and PCR data<sup>16–18</sup> suggesting a link between *M ulcerans* and water, the transmission mechanism of the disease is not known. One of the obstacles to researching this transmission mechanism is the extreme difficulty in isolating *M ulcerans* from environmental samples. The detection of the mycobacterium is usually done through PCR using the IS2404 target but, although very useful, this target might not be specific for *M ulcerans*, since it has also been found in other mycobacteria.<sup>19–21</sup> Only two pure

cultures of *M ulcerans* have ever been obtained from environmental sources.<sup>1</sup> Transmission to human beings is probably direct, through skin trauma from the water-soil reservoir, but the role of insects is increasingly suspected.<sup>16</sup> It has been demonstrated that the bite of infected naucoridae (aquatic predator insects) can transmit the disease to laboratory mice.<sup>22</sup> Because *M ulcerans* has been detected by PCR in different organisms from the aquatic ecosystem, different experimental models of transmission with passive intermediary hosts involving algae, naucoridae, aquatic snails, and fish have recently been suggested.<sup>17,22,23</sup> *M ulcerans* has also been detected by PCR in mosquitoes in Australia, but their role in transmission has still to be demonstrated.<sup>18</sup> Migratory birds might be involved in the dissemination of the disease between wetlands, leading to the appearance of new foci.<sup>17</sup>

Different factors might influence the clinical presentation of the disease—eg, the site of infection, the size of inoculum, the nature of the transmitting agent, the history of a trauma, and also the immune response of the host. An immune response mediated by Th1 lymphocytes is protective against Buruli ulcer disease but Th2 responses are not. It has been suggested that coinfection with schistosomiasis could predispose individuals infected with *M ulcerans* to express the disease by a shift to the Th2 immune response but no association has been found between the two diseases.<sup>24,25</sup> Strong IgM antibody responses to *M ulcerans* seem more frequent in patients with active Buruli ulcer than in healthy members of their families.<sup>26</sup> HIV infection

has not been reported as a risk factor for Buruli ulcer<sup>27</sup> but severe clinical forms of the disease have been described in HIV-positive patients.<sup>28</sup> In three HIV-positive Buruli ulcer patients in Akoloninga, Cameroon, we observed that the healing process improved when the patients started antiretroviral treatment (EC, unpublished data).

### Case detection and diagnosis

The burden of Buruli ulcer disease (ie, costs, extensive surgery, long hospitalisation, and development of debilitating sequelae) is mainly due to the late detection of cases. Patients often seek medical treatment when experiencing the severe and advanced stages of the disease. Late presentation is caused by several factors: (1) the disease itself—nodules can go unnoticed and when the ulcer appears, it progresses slowly, is painless, and the patient does not have systemic symptoms; (2) beliefs and stigma regarding the origin of the disease (namely witchcraft) making affected individuals reluctant to seek help; and (3) limited geographic, financial, and cultural access to medical care.

Although the clinical diagnosis of the ulcerative form is straightforward, it is more difficult for the nodule, plaque, and oedematous forms. The differential diagnosis includes pyogenic abscess in nodules, erysipelas in plaques, and cellulites in acute oedematous forms of the disease.<sup>29</sup> In endemic areas, every suspicious lesion should be treated as an *M ulcerans* infection until proven otherwise.

Tests for Buruli ulcer diagnosis include the following methods: (1) direct smear examination with Ziehl-Neelsen staining to detect the presence of acid-fast bacilli in a smear done from a swab or a biopsy; (2) culture on Löwenstein-Jensen medium at 32°C (however, this method is difficult, expensive, has a low sensitivity, and results are available only after 6–12 weeks); (3) histopathology; and (4) PCR, which allows quick detection, and is sufficiently specific in

patient samples because other environmental mycobacterium will not be present in patient samples as they are in environmental samples, the IS2404 target is sufficiently specific.

The sensitivity of these diagnostic tests<sup>30</sup> is estimated to be 40–80% for Ziehl-Neelsen staining, 20–60% for culture, over 90% for histopathology, and over 90% for PCR. In practice, however, the diagnosis of Buruli ulcer is based on clinical aspects and rarely confirmed because of limited access to laboratory services. Ziehl-Neelsen staining is the only method that could be easily implemented in the field. PCR is the quickest and most sensitive diagnostic tool but, unfortunately, presents technical difficulties (eg, cold chain requirement, stable electric supply and qualified laboratory staff) for field implementation. This technical issue could be overcome by the recent development of a dry reagent-based PCR.<sup>31</sup>

According to WHO, confirmed cases require two positive results among the diagnostic tests,<sup>30</sup> but WHO considers the possibility that one positive result might be sufficient in a context of a high clinical suspicion.<sup>32</sup>

### Treatment

Surgery remains the main treatment for Buruli ulcer. Wide excision margins, including healthy tissues, are recommended to stop the infection and prevent recurrence or relapse at the same site. Surgery in the early stages of infection is curative and highly cost-effective, since it requires a simple excision followed by an immediate closure. However, in the disease's later stages, wide and traumatising excisions are needed, followed by skin grafting, and long hospital stays. Physiotherapy after surgery might help to prevent functional disabilities. To avoid or limit the surgical excision, different medical approaches have been explored—eg, topical treatments, hyperbaric oxygen therapy, and antibiotic treatments (panel).

#### Topical treatments

Some authors have suggested that nitrogen oxides, at a concentration of 6%, could kill *M ulcerans* in Buruli ulcer lesions quickly, as it has been shown in vitro.<sup>33</sup> A small double-blind randomised controlled trial testing the daily application of a cream releasing nitrogen oxides on ulcers of less than 15 cm in diameter for at least 6 weeks has been done in Ghana.<sup>34</sup> It showed a significant reduction of the size of the ulcer over time in the intervention group ( $p=0.03$ ). However, those results should be interpreted with caution since the two groups were not comparable and only four patients had laboratory confirmed disease. Further trials are needed to define the optimum frequency and duration of application.

In a case series<sup>35</sup> phenytoin powder appeared promising in accelerating the healing process of ulcerative Buruli ulcer lesions and decreasing the risk of hypertrophic scarring. In a small randomised controlled

#### Panel: Medical treatments

##### (A) Topical treatments

###### Chemical

- Hypochlorite
- Chlorhexidine
- Iodine derivated
- Nitrites (under evaluation)
- Phenytoin powder (under evaluation)
- Clay (under evaluation)

###### Physical

- Heat

##### (B) Systemic treatments (under evaluation)

- Hyperbaric oxygen
- Heparin
- Antibiotics

trial,<sup>36</sup> ulcer surface reduction of more than 50% was observed more frequently in patients treated with phenytoin powder (72%) compared with placebo (35%), especially in young people (<30 years) and ulcers of less than 30 cm in diameter. It has also been suggested that clay allows a quick resolution of oedema, a vigorous but non-aggressive debriding of ulcers, and accelerates healing without surgery other than skin grafting.<sup>37</sup>

Local heat at 40°C, applied to eight patients with ulcers of less than 10 cm diameter in Zaire,<sup>38</sup> led to complete healing without surgery (except for the initial

removal of necrotic tissues) and no relapse at 22 months of follow-up. However, the practicality of this treatment in remote areas (high costs, minimum infrastructure such as electricity, and total confinement to bed for at least 2 weeks) is limited.

### Systemic treatment

Although oxygen therapy seems to be ineffective,<sup>39</sup> it is believed that antibiotic treatments, eventually combined with heparin, can improve the outcome of Buruli ulcer disease.

Reference	Treatment	Objective	Lesion	Study design	Number of patients	Lab result	Follow-up	Main results	Comments
Etuaful et al <sup>40</sup>	Rifampicin plus streptomycin for 0, 2, 4, 8, or 12 weeks	Culture conversion	Nodule/plaque	Randomised controlled trial	21	Yes	12 weeks	Conversion reached after a minimum of 4 weeks	Small sample size
Kanga et al <sup>41,42</sup>	Dressing plus tri-therapy (rifampicin, amikacin, and heparin) for 3 months vs dressing only	Prevent surgery	Ulcers	Randomised controlled trial	50	Yes	3 months	Prevents surgery in 80% of patients vs 0%	Further study needed to evaluate heparin effect on antibiotic penetration
Revill et al <sup>43</sup>	Clofazimine until 1 month after complete clinical healing (3–6 months)	Prevent or limit surgery	Any	Randomised controlled trial	105	Yes	Median of 32 months (17–40)	No benefit	
Espey et al <sup>44</sup>	Rifampicin plus dapsone for 2 months	Prevent surgery	Ulcers (5–26 cm <sup>2</sup> )	Randomised controlled trial	30	Yes	2 months	No evidence of benefit	Small sample size. Further study needed
Fehr et al <sup>45</sup>	Co-trimoxazole	Prevent repetitive excisions	Ulcers of any size	Randomised controlled trial	12	Yes	14–77 days	No evidence of benefit	Small sample size. Further study needed
Klutse et al <sup>36</sup>	Phenytoin powder for 8 weeks	Prevent surgery	Ulcers (all sizes)	Randomised controlled trial	51	No	8 weeks	40% ulcers healed in the intervention group compared with 8% in the placebo group	Conference abstract, many details missing
Phillips et al <sup>34</sup>	Topical nitrogen oxides for 12 weeks	Avoid surgery	Ulcers <15 cm in diameter	Randomised controlled trial double blind pilot study	30	Yes	12 weeks	Greater reduction of the ulcer surface in intervention group compared with the placebo group	Small sample size. Intervention group older, with ulcer of longer duration and bigger surface
Darie et al <sup>46</sup>	Rifampicin, streptomycin, co-trimoxazole, ofloxacin, cephalosporin, penicillin A/M, metronidazol, or minocyclin for 1 month	Decrease failure rate	Ulcers	Observational	88	No	6 months	Only streptomycin is associated with an acceptable failure rate	
Meyers et al <sup>38</sup>	Local heat (19–74 days)	Accelerate healing after surgery	Ulcers of <10 cm	Observational	8	Histo-pathology	22 months	Healing	
Brunet de Cours-sou <sup>37</sup>	Clay	Avoid surgery	All forms	Observational	..	No	..	Healing for small lesions, skin graft only for extended lesions	

..=not reported.

**Table 1: Overview of medical treatments for Buruli ulcer**

Many antimicrobial agents have excellent in-vitro activity against *M ulcerans* and different combinations inhibit growth in animals, but evidence in the treatment of Buruli ulcer in human beings is only anecdotal. Very few randomised controlled trials have been done and the existing ones are too small and/or not conclusive. The difficulty of demonstrating the beneficial effects of antibiotics might be caused by treatment effects being undetected because of irreversible tissue damage and necrosis, or because of poor irrigation in necrotic tissues hampering the penetration of the drugs into the tissues where *M ulcerans* remains. It is also possible that ongoing necrosis is caused by persistent toxin load rather than active mycobacterial infection. Several antimicrobial agents have been tested in the treatment of Buruli ulcer as monotherapy or bitherapy (table 1).

The use of monotherapies is generally ineffective. Clofazimine, an anti-leprosy drug evaluated in a large randomised controlled trial,<sup>43</sup> did not improve the healing process, nor reduce the number of surgical excisions, nor prevent recurrences. A randomised controlled trial of co-trimoxazole (trimethoprim-sulfamethoxazole)<sup>45</sup> did not show any benefits and was not conclusive because of the small sample size and because the treatment groups differed with regard to initial surgical treatment and duration of follow-up. In a cases series in Côte d'Ivoire in which different antibiotics were used in monotherapy for 1 month, the lowest failure rate was associated with streptomycin.<sup>46</sup> Studies in animal models showed that aminoglycosides (eg, streptomycin and amikacin) and rifampicin have a strong bactericidal activity when used alone.<sup>47,48</sup> However, *M ulcerans* could become resistant to rifampicin if this drug is used in monotherapy.<sup>49</sup>

Based on these results and some observational studies, WHO recommended a combination of rifampicin and streptomycin for 8 weeks for the management of Buruli ulcer.<sup>50</sup> With this treatment, experts hope to reduce the indications of surgery or, at least, the extent of the surgery and hope also to decrease the relapse rate. A small clinical trial showed that a minimum of 4 weeks of this combination inhibits the growth of *M ulcerans* in nodules and plaques, confirmed by PCR and/or acid-

fast bacilli examination and/or culture.<sup>40</sup> However, the size of this trial is too small to conclude effectiveness of this treatment and its efficacy in preventing relapse or dissemination has not yet been evaluated.

It has been suggested that heparin combined with rifampicin and aminoglycoside treatment could improve blood circulation and antibiotic penetration. A case study report showed excellent results of this combination in a young girl who could not be operated on because of the location of the lesion on the face.<sup>41</sup> This observation has been followed by a non-blinded randomised controlled trial comparing local nursing alone with local nursing plus the tri-therapy of heparin, rifampicin, and amikacin.<sup>42</sup> In the group receiving only dressing care, 100% of the patients needed surgery, whereas in the group receiving the tri-therapy, only 20% had to go through surgery. However, the design of the study makes it difficult to assess the role of heparin versus that of the antibiotics.

The combination of clarithromycin and rifampicin is regularly used as part of standard treatment protocols in Australia<sup>51</sup> but its efficacy has never been evaluated.

In a randomised controlled trial done in Côte d'Ivoire,<sup>44</sup> the combination of rifampicin and dapsone showed ulcer size reduction in the treatment group compared with the placebo group but these results are inconclusive due to the small sample size and the imbalances of the ulcer size at baseline.

Some antimicrobial agents have shown promising results in vitro and in animal models (table 2) but clinical trials are needed to assess their effectiveness in human beings. In vitro, *M ulcerans* is highly susceptible to fluoroquinolones<sup>52</sup> but in mice, their effect is bacteriostatic.<sup>49</sup> Sitafoxacin, a new fluoroquinolone, has been demonstrated to be superior to other fluoroquinolones both in vitro and in vivo, especially when combined with rifampicin.<sup>53,54</sup> The combination of rifampicin, clarithromycin, and sparfloxacin has been tested in mice but it is not as effective as rifampicin with an aminoglycoside.<sup>49,55,56</sup> Rifalazil (benzoxazino-rifamycin or KRM-1648), derived from rifampicin, seems to have a better inhibitory activity than rifampicin in mice.<sup>57</sup> In-vitro studies showed that epiroprim, a dihydrofolate reductase inhibitor, has a good inhibitory activity on the growth of *M ulcerans* and that there is a strong synergic activity against *M ulcerans*<sup>58</sup> when dapsone and epiroprim are combined. At least one new drug (a diarylquinoline known as R207910) has a remarkable activity in vitro against many mycobacterial species, including *M tuberculosis* and *M ulcerans*.<sup>59</sup>

Overall, no antibiotic combination has proven effective in the management of Buruli ulcer up to now. Despite a lack of evidence, it is believed that antimycobacterials in the right combination for the appropriate duration may reduce the healing time for ulcers and the recurrence rate, and might help avoid surgery or at least limit its extent. Rifampicin seems to

Antibiotic(s)	Study phase	Remarks
Fluoroquinolones	In vitro/mice	Bacteriostatic. Some fluoroquinolones (eg, sparfloxacin, moxifloxacin, amifloxacin, and especially sitafoxacin) seem to work better than others, at least in vitro
Rifalazil	Mice	Inhibitory activity greater than rifampicin. Bactericidal towards <i>M ulcerans</i>
Epiroprim alone <sup>58</sup>	In vitro	Epiroprim > brodimoprim and K130 > trimethoprim (the last being totally ineffective even at high concentration)
Epiroprim plus dapsone <sup>58</sup>	In vitro	Epiroprim plus dapsone: synergic action Brodimoprim plus dapsone: mainly additive effects Mouse results suggest that rifampicin plus epiroprim plus dapsone could be beneficial in the clinical treatment of advanced ulcers
Diarylquinoline	In vitro	Remarkable activity

**Table 2: Overview of potential antibiotic treatments for Buruli ulcer disease**

be the key antibiotic for the treatment of Buruli ulcer but needs to be combined with another antibiotic to increase its effect and prevent the development of resistance. The choice between aminoglycosides and fluoroquinolones is still debated among Buruli experts, even though WHO issued a provisional guidance in 2004 recommending the combination of rifampicin and streptomycin for 8 weeks.<sup>47</sup> However, the long-term side-effects of aminoglycosides are quite substantial, and their effectiveness has not yet been formally demonstrated by well conducted clinical trials. Moreover, they are contraindicated in pregnant women and the injectable pathway is not ideal in the context of the HIV epidemic in sub-Saharan Africa. Fluoroquinolones do not have the drawbacks of the aminoglycosides and have good bone penetration. However, these drugs are contraindicated in sun exposure, in growing children, and in pregnant and breast-feeding women. Another important concern is that rifampicin and streptomycin are effective antituberculosis drugs. Their use in Buruli ulcer, without excluding concomitant active tuberculosis, might increase the risk of developing resistant tuberculosis, which could become a real threat if Buruli ulcer disease continues to progress as it has over the past 20 years. In addition, the progression of the HIV epidemic makes it more likely that more Buruli ulcer patients will be coinfecting with tuberculosis.

### Control strategies

To date, public-health efforts promoting early detection and rapid treatment (by active case finding and health education) have achieved the best results in terms of morbidity and costs of the disease.<sup>7,60</sup> However, unlike tuberculosis, early detection and treatment will not decrease transmission, as human-to-human transmission is very rare. In a focus in southern Australia, limitation of irrigation from a contaminated water

source has been associated with a dramatic reduction of new Buruli cases.<sup>61</sup>

A randomised controlled trial done in 1976 in Uganda<sup>62</sup> provided evidence of a 47% short-term protective effect (6 months to 1 year) of BCG vaccination against Buruli ulcer. The trial did not determine whether BCG offered long-lasting protection. Two case series from Benin<sup>63</sup> and Cameroon<sup>64</sup> suggested that BCG vaccination could protect children against severe forms of the disease (osteomyelitis and multiple lesions).

Inexpensive prevention strategies such as wearing protective clothing when farming and the immediate cleansing of traumatic skin injuries might also help.<sup>26,65,66</sup>

### Future perspectives

The discovery of the plasmid harbouring the genes responsible for mycolactone production and the sequencing of the complete genome of *M ulcerans*<sup>67</sup> together provide a fundamental resource for researchers. The identification of *M ulcerans*-specific antigens might help to develop a genetic vaccine. The knowledge of the genome will reveal enzymes involved in pathogenesis and potential virulence factors, which might promote the development of new drugs targeting these specific pathways. A better understanding of the molecular epidemiology might help to discover how the disease is transmitted.

Where do we go from here? Table 3 summarises the current situation and future research needs. More general data are required regarding the incidence and risk factors for Buruli ulcer disease.

Until the mode of transmission is fully understood, general environmental measures to protect communities cannot be defined. If the role of aquatic bugs in transmission or as intermediary hosts is confirmed, preventive environmental measures such as the ones used for schistosomiasis could be applied—eg,

	Current situation, main problems	Research and development needs
Burden of disease	Emerging disease due to environmental factors Under-reporting of the disease through passive surveillance system Huge socioeconomic impact in the most affected communities mainly due to late diagnosis High recurrence rate	National prevalence surveys for identifying new foci and mapping Develop strategies for active case finding Make Buruli ulcer a nationally reportable disease in affected areas Improve reporting system through the existing community-based surveillance system
Control of the disease	Mode of transmission not well understood Eradication of the suspected water points impossible, especially in the most endemic countries of Africa	Identify the vectors to develop eventual vector-control strategies Test health education and individual protective measures
Prevention of disease	Except BCG, which confers only a very small protection, no vaccine available	Development of an affordable and effective vaccine as a long-term objective
Diagnosis	Clinical diagnosis difficult at early stages Invasive procedure required for sample collection at early stages PCR, the most sensitive tool for confirmation, is expensive and difficult to implement in the field	Produce affordable, rapid, and non-invasive diagnostic tests that can detect the early stages of the disease and are easy to implement in the field Promote outreach activities and health education strategies in endemic areas for early case detection
Treatment	In the late stages of the disease, the treatment requires mutilating and expensive surgery and long hospitalisation No antibiotic has been formally proven to be effective The antibiotic treatment recommended by WHO has side-effects	Field-standardised measures to identify patients coinfecting with tuberculosis Identify new combination therapies specific against <i>M ulcerans</i> and tested on human patients

Table 3: Summary of the current situation and needs

### Search strategy and selection criteria

We identified information for this review through a PubMed search (English and French) up to October 2005, and references from relevant articles together with other published data of the WHO website and unpublished data presented at annual WHO advisory group meetings on Buruli ulcer. Search terms were "Buruli" OR "*Mycobacterium ulcerans*".

improved irrigation and agriculture practices (eg, snail habitats could be reduced by removing vegetation, by draining and filling, or by lining canals with concrete, and treating snail-breeding sites with molluscicides). Draining the water sources of *M ulcerans* is rarely applicable in the rural communities of Africa, unless access to other, safer water sources are offered to these populations. The effectiveness of individual protective measures such as wearing clothes that cover the extremities or rubber boots when farming needs to be explored further.

The need for vaccination in endemic areas is clear. Because of the limited role of BCG, a new vaccine is urgently needed for the prevention of Buruli ulcer. Among the prospects for developing a new vaccine, non-virulent *M ulcerans* strains have been identified and could be used for developing new vaccines.<sup>68</sup> Also, the sequencing of the *M ulcerans* genome could help in identifying specific antigens from which a plasmid DNA genetic vaccine could be developed.<sup>69,70</sup>

Now that the combination of rifampicin and streptomycin recommended by WHO has been implemented in several Buruli ulcer endemic countries (eg, Benin, Cameroon), it is difficult to do randomised controlled trials to determine the effectiveness of this approach. Therefore, it is essential to ensure close monitoring and data collection for all Buruli ulcer cases treated with the WHO recommended treatment, describing precisely the treatment schemes in use, the results regarding the localisation and clinical form of Buruli ulcer, reduction of the ulcer size or oedema, speed of healing, relapse rate, side-effects, impact on the extension of the surgery, and long-term impact on tuberculosis (ie, the development of resistant forms). Additionally, feasibility and cost-effectiveness should be evaluated. Sitaflaxacin, rifalazil, epiroprim, and the diarylquinolone R207910 should be tested in combination with other antimicrobial agents. Pharmacokinetic and/or pharmacodynamic modelling needs to be considered before clinical studies are planned, so that appropriate human doses are used. Meanwhile, there is an urgent need to identify cheap, safe, and effective oral combinations of antibiotics. There is also a need to properly assess the effects of heparin on antibiotic penetration with studies that compare groups of patients on antibiotic treatment with or without heparin.

Also, because the necrotic lesions are caused by the production of mycolactone, further work should look at the possibility of developing new drugs aimed at inhibiting the toxin or its production.

PCR is a quick and sensitive test for detecting *M ulcerans*, and the new dry reagent form would make it easier to use in practice. However, it is very expensive and requires punch biopsies for the non-ulcerative stage of the disease. It would be desirable to have rapid and non-invasive diagnostic tests that can be carried out in the field. There are hopes for serodiagnostic assays for early detection based on *M ulcerans*-specific antigens.<sup>1</sup> However, it is uncertain how specific these tests will be in areas where all the members of affected communities are exposed to this environmental pathogen. Also, operational research is required to test the effectiveness and applicability of these new tests in remote villages with little infrastructure. Detection strategies will need to define the target groups, the optimal screening frequency, and how to access the most affected populations. The added value of these tests compared with clinical examination in endemic areas might be questionable when the easiest and quickest treatment of most of the nodules is excision.

Buruli ulcer was declared an emerging disease in 1998. The WHO Department of Control of Neglected Tropical Diseases has recently created a Technical Advisory Group on Buruli ulcer, which will provide advice to WHO and its partners on all aspects of Buruli ulcer control and research. Buruli experts have invested much effort in research. The recent sequencing of the *M ulcerans* genome opens new prospects and hope for the future of Buruli ulcer management. However, these prospects are still far in the future, and such hopes should not obscure the fact that when the disease is tackled in the early stages, treatment is extremely simple and cost-effective. Efforts should be directed towards developing strategies for early diagnosis and treatment of the disease, including health education for the most affected populations to increase awareness and change misguided beliefs about the disease, training of health-care workers on early detection and minor surgery, and the implementation of outreach activities (eg, active case finding, health education) in the most affected areas.

### Conflicts of interest

We declare that we have no conflicts of interest.

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