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Increasing Experience with Primary Oral Medical Therapy for *Mycobacterium ulcerans* Disease in an Australian Cohort

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Buruli ulcer (BU) is a necrotizing infection of subcutaneous tissue that is caused by *Mycobacterium ulcerans* and is responsible for disfiguring skin lesions. The disease is endemic to specific geographic regions in the state of Victoria in southeastern Australia. Growing evidence of the effectiveness of antibiotic therapy for *M. ulcerans* disease has evolved our practice to the use of primarily oral medical therapy. An observational cohort study was performed on all confirmed *M. ulcerans* cases treated with primary rifampin-based medical therapy at Barwon Health between October 2010 and December 2014 and receiving 12 months of follow-up. One hundred thirty-two patients were managed with primary medical therapy. The median age of patients was 49 years, and nearly 10% had diabetes mellitus. Lesions were ulcerative in 83.3% of patients and at WHO stage 1 in 78.8% of patients. The median duration of therapy was 56 days, with 22 patients (16.7%) completing fewer than 56 days of antimicrobial treatment. Antibiotic-associated complications requiring cessation of one or more antibiotics occurred in 21 (15.9%) patients. Limited surgical debridement was performed on 30 of these medically managed patients (22.7%). Cure was achieved, with healing within 12 months, in 131 of 132 patients (99.2%), and cosmetic outcomes were excellent. Primary rifampin-based oral medical therapy for *M. ulcerans* disease, combined with either clarithromycin or a fluoroquinolone, has an excellent rate of cure and an acceptable toxicity profile in Australian patients. We advocate for further research to determine the optimal and safest minimum duration of medical therapy for BU.

Buruli ulcer (BU) is a necrotizing infection of subcutaneous tissue that is caused by *Mycobacterium ulcerans* and is responsible for disfiguring skin lesions (1). While the disease is most common in West African countries (2), it is also endemic to specific geographic regions in the state of Victoria in southeastern Australia (3). Within the Bellarine Peninsula region of Victoria, BU is generally most common among older adults (3), in contrast to the affected demographic in West Africa, which is predominantly children (4).

Antibiotics have been shown to be highly effective at sterilizing lesions and preventing disease recurrences when used alone or in combination with surgery (1, 5–9). Mounting evidence of the effectiveness of antibiotic therapy for *M. ulcerans* disease has substantially shifted the balance in favor of medical therapy over surgery, and our practice has evolved to using primarily oral medical therapy (1, 5). In 2013, we described the results of medical therapy in a group of 43 patients, among whom 42 patients (98%) healed without recurrence within 12 months (1). Here we provide details of our increasing experience with the use of primary oral medical therapy in a large observational Australian cohort.

MATERIALS AND METHODS

This is an observational cohort study approved by Barwon Health's Human Research and Ethics Committee. All previously collected medical data were analyzed anonymously.

Data on all confirmed *M. ulcerans* cases managed at Barwon Health have been collected prospectively since January 1998. From October 2010, our standard treatment practice for initial *M. ulcerans* lesions has comprised combination antimicrobial therapy, with limited surgical debridement performed to aid wound healing, unless antibiotics are declined or contraindicated (10). The following data were extracted from medical records: patient demographics, comorbid conditions, size of the BU lesion(s), date of diagnosis, antimicrobial regimen and duration, and details of any adjunctive surgical procedures. Cases treated between October 2010 and December 2014 and receiving 12 months of follow-up were included in this study.

Definitions. An *M. ulcerans* case was defined as previously described (1). In brief, cases had the presence of a lesion clinically suggestive of *M. ulcerans* plus a positive culture, positive PCR, and/or consistent histopathology for *M. ulcerans*.

Primary medical treatment was defined as treatment of an *M. ulcerans* lesion with either antimicrobials alone or antimicrobials in conjunction with limited surgical debridement. Drugs and dosages for adults included ciprofloxacin at 500 mg twice daily, moxifloxacin at 400 mg daily, rifampin at 10 mg/kg of body weight/day (up to a maximum of 600 mg daily), and clarithromycin at 500 mg twice daily; for children, the clarithromycin dose was 7.5 to 15 mg/kg daily in divided doses (up to a maximum of 500 mg twice daily).

Limited surgical debridement was defined as curettage of the lesion or a minor excision to remove excess granulation tissue and to debride ulcer margins, with or without the use of a split skin graft (SSG). Limited surgical debridement was undertaken primarily to remove necrotic tissue from *M. ulcerans* lesions in order to promote healing by secondary intention. Patients who underwent extensive surgery (as defined previously) (1) were excluded from this formal analysis.

Paradoxical reactions were defined by the presence of one or both of the following features: (i) an initial improvement in the clinical appearance of an *M. ulcerans* lesion upon antibiotic treatment, followed by de-

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TABLE 1 Baseline characteristics of medically treated *M. ulcerans* patients

Characteristic	Value (no. [%])[a]
Median (IQR) age (yr)	49 (23.5–72)
Male gender	75 (56.8)
WHO category	
1	104 (78.8)
2	19 (14.4)
3	9 (6.8)
Lesion type	
Ulcer	110 (83.3)
Nodule	9 (6.8)
Edematous	10 (7.6)
Plaque	3 (2.3)
Site	
Upper limb	44 (33.3)
Lower limb	86 (65.2)
Trunk	2 (0.2)
Comorbidities	
Immunosuppression	6 (4.5)
Malignancy	5 (3.8)
Diabetes mellitus	13 (9.8)

[a] Unless indicated otherwise.

terioration of the lesion or its surrounding tissues or the appearance of a new lesion(s); and (ii) examination of excised tissue from the clinical lesion showing evidence of an intense inflammatory reaction consistent with a paradoxical reaction (11).

Treatment success was defined as complete healing of the *M. ulcerans* lesion without recurrence within 12 months of treatment commencement.

Recurrence of *M. ulcerans* infection was defined as a new lesion (meeting the case definition for *M. ulcerans* appearing either in the wound, locally, or in another distant body site) due to relapse or new infection after initial medical treatment and/or surgical excision of the initial *M. ulcerans* infection. It was distinguished from a paradoxical reaction based on histopathology (as described above).

Treatment failure was defined as either failure of healing of the *M. ulcerans* lesion within 12 months or the presence of a recurrent *M. ulcerans* lesion within 12 months of treatment completion that either (i) was culture positive for *M. ulcerans* or (ii) had histopathology showing a necrotic ulcer with the presence of acid-fast bacilli, consistent with acute *M. ulcerans* infection, and did not show evidence of a paradoxical reaction.

A complication of medical therapy was defined as an adverse event attributed to an antibiotic that required cessation of that medication.

Data were collected using Epi-Info 6 (CDC, Atlanta, GA) and analyzed using STATA 12 (StataCorp, TX). Categorical values were compared using the Mantel-Haenszel test, and median values were compared using the Mann-Whitney test.

RESULTS

From 1 October 2010 through 31 December 2014, there were 160 patients with *M. ulcerans* disease managed at Barwon Health. Twenty-eight patients (17.5%) were excluded from this analysis because they underwent extensive surgical excision, either with (20 patients) or without (8 patients) adjunctive antimicrobial therapy.

Baseline characteristics can be seen in Table 1. All 132 patients were primarily managed as outpatients. The majority of patients were male (75/132 patients [56.8%]), the median age was 49 years (range, 1 to 95 years), and nearly 10% of patients had diabetes mellitus. Lesions were ulcerative in 83% of patients and at WHO stage 1 in 78.8% of patients (Table 1). One hundred thirty patients had single *M. ulcerans* lesions, one patient had 2 lesions, and one patient had 10 distinct lesions.

Antimicrobial regimens were all rifampin-based regimens. Rifampin was combined as a first-line agent with ciprofloxacin in 80 (60.6%) patients, with clarithromycin in 50 (37.9%) patients, and with moxifloxacin in 2 (1.5%) patients. The median duration of therapy was 56 days (interquartile range [IQR], 24 to 96 days). Twenty-two patients (16.7%) completed fewer than 56 days of antimicrobial treatment, and of these 22 patients, 10 completed fewer than 42 days of therapy and 5 received ≤28 days of therapy.

Antibiotic-associated complications requiring cessation of one or more antibiotics occurred in 21 (15.9%) patients. On examining the data by antibiotic regimen, 16 of 80 patients (20%) treated with rifampin and ciprofloxacin ceased one or more agents, 4 of 50 patients (8%) treated with rifampin and clarithromycin ceased one or more agents, and 1 of 2 patients treated with rifampin and moxifloxacin ceased one or more agents. On examining the data by individual antibiotic agent, rifampin was ceased in 12 patients (9.1%), ciprofloxacin was ceased in 12 patients (9.1%), clarithromycin was ceased in 5 patients (3.8%), and moxifloxacin was ceased in 1 patient. Among the entire *M. ulcerans* infection cohort, rates of discontinuation were similar between agents.

Paradoxical reactions to antimicrobial therapy occurred in 34 patients (25.7%) after a median of 48 days of antibiotics (IQR, 29 to 69 days).

Limited surgical debridement was performed on 30 of these medically managed patients (22.7%). Debridement was performed on 18 patients, and an SSG was required in 12 patients for coverage of the defect. For the 30 patients who underwent debridement, 16 had cultures performed on surgical specimens: 8 specimens were collected before treatment, and 8 specimens were collected after treatment had been completed. All 8 specimens obtained after treatment were culture negative.

Cure was achieved, with healing within 12 months, in 131 of 132 patients (99.2%), and cosmetic outcomes were excellent for medically managed *M. ulcerans* cases.

As previously described (1), the single treatment failure was in a 17-month-old boy with a nodular lesion which enlarged, ulcerated, progressed, and failed to heal within 12 months, despite completion of primary therapy with rifampin (10 mg/kg/day) and clarithromycin (13 mg/kg/day in twice-daily dosing) in liquid formulations. We determined that the possible reasons for primary treatment failure are as follows: the nonulcerative nature of the lesion, which may have been more difficult to sterilize; the syrup preparations of antimicrobials, with uncertain rifampin bioavailability; and interactions between rifampin and clarithromycin leading to unintentional underdosing in the absence of drug levels being determined.

DISCUSSION

We have described our experience with primary medical management of *M. ulcerans* disease in southeastern Australia by use of oral rifampin-based regimens. We demonstrated the effectiveness of this treatment approach with a 99% cure rate at 12 months for a large cohort of 132 patients. The toxicity profile is acceptable, and cosmetic results are excellent. In our practice, the criteria for ex-

clusive medical therapy are based primarily on patient willingness to take antimicrobials and no contraindications to antimicrobial therapy (such as drug interactions or severe hepatic disease). Lesions of all sizes are eligible for primary oral medical therapy, although healing may be slow for WHO category 2 and 3 lesions, occurring anywhere from 11 to 15.5 weeks (7) to 30 weeks (8) after initiation of antimicrobials. In our cohort, over 20% of patients had WHO category 2 and 3 lesions, and they healed successfully within 12 months. Limited surgery, such as debridement, was employed in about one-fifth of the members of our medically managed cohort, and the easy access of patients to this type of adjunctive treatment can be important for promoting healing of *M. ulcerans* disease.

Treatment with medical therapy alone for *M. ulcerans* disease gained momentum in 2004, when rifampin and streptomycin were recommended by the WHO on the basis of mouse models and limited human data (12, 13). In 2007, Chauty et al. demonstrated the effectiveness of 8 weeks of a rifampin-streptomycin (RS8) combination in Benin (14), and success with this regimen was again confirmed by Sarfo et al. in Ghana in 2010 (7). At Barwon Health, we demonstrated in 2007 in Australian cohorts that antibiotics including regimens combining oral rifampin and fluoroquinolones were effective at curing lesions and preventing recurrences when combined with surgical excision. Furthermore, in 2010, a randomized controlled trial in Ghana was published that found RS8 to have an efficacy similar to that of 4 weeks of rifampin-streptomycin followed by 4 weeks of rifampin-clarithromycin (RC) (RS4RC4) (8). These successes paved the way for the use of the fully oral combinations of rifampin and fluoroquinolones (RFQ) and RC in both Australia and Benin (1, 5, 9). Most recently, treatment success was demonstrated in Ghana with an even shorter duration of injectable streptomycin therapy of 2 weeks (RS2) followed by oral RC for 6 weeks (4). Results to be obtained from the WHO-led trial comparing the efficacies of RC and RS in Ghana and Benin are likely to aid in the development of a definitive treatment policy (https://clinicaltrials.gov/ct2/show/NCT01659437).

In our study, 12 patients ceased therapy early because of side effects. This rate of adverse effects is in keeping with rates previously described for rifampin, clarithromycin, and ciprofloxacin in our experience (1, 5, 15), which are higher than rates reported for younger populations in Africa (7). The older median age of our cohort likely contributes significantly to reduced drug tolerability in our cohort (15). An analysis of the duration of therapy completed among our cohort revealed that 16% of patients completed fewer than 56 days (8 weeks) of therapy. Among these patients, 15 patients completed fewer than 42 days (6 weeks) of therapy, without treatment failure. We previously determined that the success rate is 100% if patients complete >28 days of therapy (15). We believe that it is possible that therapy may be shortened to 4 to 6 weeks in many cases, although more research in this field is required.

In the Australian experience, injectable therapy was not a favorable therapeutic option because of the lack of availability of streptomycin, the fact that most patients are outpatients, and concerns about toxicity and the need for monitoring for adverse events, especially in our more elderly population (6). Therefore, both experience and confidence have evolved with the use of oral rifampin-based therapy regimens for *M. ulcerans* disease. In our practice, rifampin is combined with either clarithromycin or a fluoroquinolone. Rifampin dosing is uniform internationally; however, in the case of clarithromycin, previous studies have used different doses, ranging from 7.5 mg/kg daily (4, 7) to 12 mg/kg daily (9). While the optimal dose of clarithromycin that should be prescribed is not known, there is a considerable risk of an interaction with rifampin which induces the hepatic cytochrome P-450 system, inducing the metabolism of clarithromycin (16). Wallace and others demonstrated for patients with *Mycobacterium avium* complex infection treated with RC that the mean serum level of clarithromycin given as a single agent was 5.4 ± 2.1 μg/ml but that this decreased substantially, to 0.7 ± 0.6 μg/ml, in patients receiving rifampin with clarithromycin (16). The pharmacokinetics of the RC combination (with clarithromycin dosed at 7.5 mg/kg/day) for *M. ulcerans* infection were studied, and the results indicated that the median concentration of clarithromycin was above the MIC for *M. ulcerans* but that the concentration of 4-hydroxyclarithromycin was not (17). The authors of that study therefore recommended that clarithromycin should be dosed at 7.5 mg/kg twice daily (or 15 mg/kg daily in an extended-release formulation) to ensure higher levels of exposure to clarithromycin and an increase in the time above the MIC compared to those achieved with a dose of 7.5 mg/kg once daily (17). We concur with this recommendation in order to avoid subtherapeutic treatment with clarithromycin, and hence secondary rifampin resistance development as may occur when rifampin monotherapy is used (18).

Conclusions. Primary rifampin-based oral medical therapy for *M. ulcerans* disease, combined with either clarithromycin or a fluoroquinolone, has an excellent rate of cure and an acceptable toxicity profile in Australian patients. We advocate for further research to determine the optimal and safest minimum duration of medical therapy for BU.

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