"Paradoxical" immune-mediated reactions to *Mycobacterium ulcerans* during antibiotic treatment: a result of treatment success, not failure

Daniel P O'Brien, Michael E Robson, Peter P Callan and Anthony H McDonald

We present the first clinical descriptions of immune-mediated paradoxical reactions to effective antibiotic treatment for Mycobacterium ulcerans *infection, which result in clinical deterioration after initial improvement. Recognition of this phenomenon could prevent unnecessary changes to antibiotic regimens, and might obviate the need for, or reduce the extent of, further surgery.*
(MJA 2009; 191: 564-566)

Clinical records

Patient 1

A 46-year-old man presented with a 3-month history of a slowly enlarging asymptomatic lesion on the lateral aspect of his right leg, that ulcerated and reached a size of 1.5 cm in diameter. A diagnosis of *Mycobacterium ulcerans* infection was confirmed by histopathological examination, which showed the classic appearance of a necrotic lesion involving the dermis and subcutaneous tissue, with a sparse acute inflammatory reaction, fat cell ghosts and large numbers of extracellular acid-fast bacilli (AFB)[1] (Box, A and Box, B). In addition, culture and polymerase chain reaction (PCR) tests of excised tissue were positive for *M. ulcerans*.

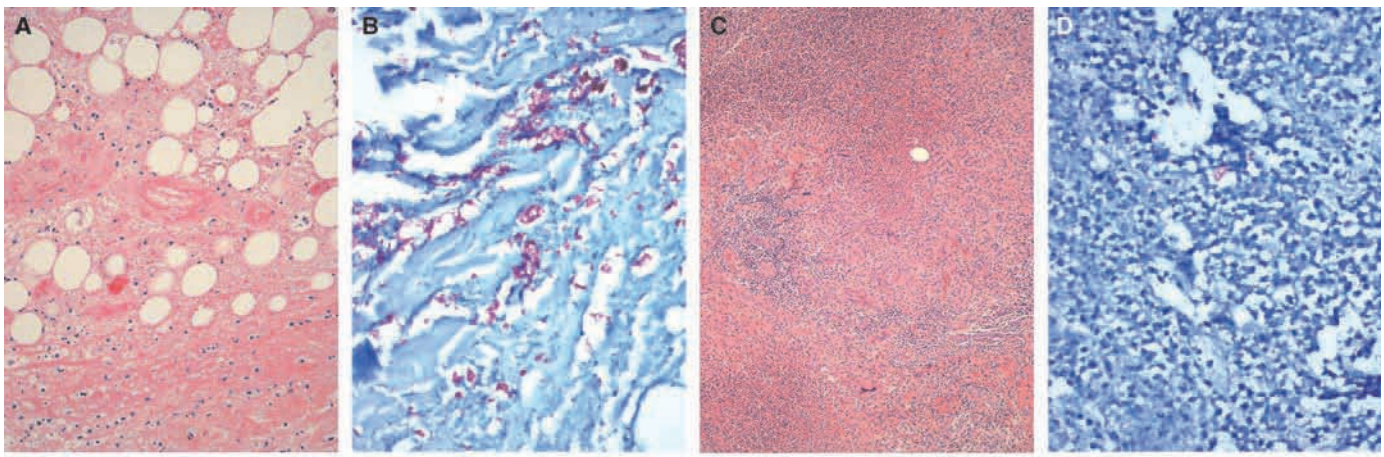
The ulcer was excised with primary skin closure and clear histological margins, and adjunctive antibiotic therapy (with 300 mg rifampicin and 500 mg ciprofloxacin, twice daily) was commenced with a planned duration of 3 months. This lesion had not recurred in the ensuing 7 months.

However, in the 2 days before the first lesion was treated, the patient spontaneously developed, proximally, an area of erythema and swelling over the right knee. Although secondary *M. ulcerans* infection at the site was considered, bacterial cellulitis or bursitis was possible, so it was decided to observe the response to intravenous therapy with cephazolin. Initially, the amount of induration and erythema decreased significantly, but did not resolve completely.

About 5 weeks later, the area became increasingly inflamed and, over the following month, became fluctuant but not ulcerative. An aspirate of the lesion confirmed the presence of *M. ulcerans* with AFB seen on microscopy, and a positive *M. ulcerans* PCR. The patient, therefore, underwent further wide excision of the lesion with a split-thickness skin graft. The antibiotic regimen was changed by replacing ciprofloxacin with clarithromycin (500 mg twice daily) while continuing with rifampicin.

Histopathological examination of the excised tissue showed extensive undermining necrosis in the fat and subcutaneous tissue, accompanied by a florid mixed inflammatory reaction with multinucleated giant cells (Box, C). However, only a few sparse AFB were seen (Box, D). Mycobacterial cultures of the aspirate and the excised tissue were negative at two separate laboratories. The

Lesions in Patient 1



First lesion, before antibiotic treatment showing: ***(A)*** *a sparse acute inflammatory reaction around necrotic fat and subcutaneous tissue (haematoxylin and eosin stain; original magnification, ×100; and* ***(B)*** *high numbers of extracellular acid-fast bacilli (pink) on Wade–Fite stain (original magnification, ×400).*

Second lesion after 11 weeks of antibiotic treatment showing: ***(C)*** *a dense inflammatory reaction including multinucleated giant cells (haematoxylin and eosin stain; original magnification, ×100; and* ***(D)*** *only one small cluster of extracellular acid-fast bacilli on Wade–Fite stain (original magnification, ×400).*

patient completed a further 7 weeks of postoperative antibiotic therapy and, after 6 months, there was no evidence of further *M. ulcerans* lesions.

Patient 2

A 66-year-old man presented with a 6-week history of a painless ulcer on the left elbow, with significant surrounding induration. A diagnosis of *M. ulcerans* infection was made, and antibiotic therapy (300 mg rifampicin and 500 mg ciprofloxacin, twice daily) was commenced. One week later, a wide excision and split-thickness skin graft was performed, although not all the indurated area was excised. Culture and PCR of excised tissue was positive for *M. ulcerans*. Histopathological examination of the excised lesion showed classic features of *M. ulcerans* infection, with necrosis of the dermis and subcutaneous tissue, a limited and focal inflammatory response, and large numbers of extracellular AFB. These changes extended to the deep excision margin.

The clinical appearance of the indurated area that was not excised improved significantly after surgery. However, 6 weeks later and while still taking the antibiotics, over 48 hours, the patient developed erythema and induration of the left elbow region that extended inferiorly from the lower edge of the excisional wound onto the forearm; the condition had the clinical appearance of cellulitis.

This was thought to be a secondary bacterial infection, but it worsened despite intravenous therapy with cephazolin. The patient, therefore, underwent further operative exploration, during which extensive liquefied fat extending distally into the forearm was found and debrided. Histopathological examination of excised tissue showed inflamed granulation tissue with lymphocytes, histiocytes and occasional multinucleated giant cells, and scattered necrotic foci. There were no AFB detected on microscopy and, although PCR of the tissue was positive, cultures were negative for *M. ulcerans*. The antibiotic regimen was changed by replacing rifampicin with clarithromycin (500 mg twice daily), and the patient took clarithromycin for a further 3 months and ciprofloxacin for a further month. The wound healed completely by secondary intention, and there has been no evidence of recurrence of *M. ulcerans* infection 10 months after the second operation.

Discussion

A "paradoxical" reaction describes a deteriorating response to treatment of an infection after initial improvement.[2] Such reactions have frequently been described in infections with a number of mycobacterial species, including those causing tuberculosis, avium-intracellulare complex and leprosy, and although they occur most commonly in severely immunosuppressed patients with HIV/AIDS who are undergoing antiretroviral therapy, they can occur in immunocompetent hosts.[2,3] The pathogenesis relates to an enhanced immune response to mycobacterial antigens on treatment that produces deleterious clinical effects.[1] Its immune basis means that this phenomenon is also referred to as "immune reconstitution syndrome".

M. ulcerans causes necrotising lesions of skin and subcutaneous tissue. Despite large numbers of extracellular mycobacteria, lesions are characterised by a poor inflammatory response.[4] This is probably induced by immuno-inhibitory characteristics of mycolactone, the exotoxin produced by *M. ulcerans*,[5,6] which plays a crucial role in the pathogenesis of infection with this organism. Treatment primarily involves surgical excision; however, recent evidence has shown that antibiotics can have an effective role,[7,8] and their use is now recommended.[9,10]

Clinical paradoxical reactions during the treatment of *M. ulcerans* have not been previously described. Clinical deterioration during antibiotic treatment can be interpreted as treatment failure, leading to further expensive and potentially disfiguring surgery, and a change in antibiotic regimens or a prolongation of their use. In the two cases we describe, involving treatment of *M. ulcerans* infections in patients from an endemic area in the Bellarine Peninsula of south-eastern Australia,[7] initial improvement during antibiotic therapy was followed by a paradoxical worsening in the clinical appearance. This was first interpreted as treatment failure, but we believe it was subsequently shown to result from an immune-mediated reaction to effective antibiotic treatment.

We feel that in Patient 1, the lesion on the right knee represented a secondary *M. ulcerans* infection that was present at the time the lower right leg lesion was treated. It showed initial improvement with antibiotic therapy, but there was subsequent clinical deterioration consistent with a paradoxical reaction to mycobacterial antigens at the site. Likewise, in Patient 2, the clinical postoperative deterioration at the wound margins after initial improvement in an area of incomplete excision of the mycobacterial infection represented a paradoxical reaction to remaining mycobacterial antigens. To our knowledge, these are the first clinical case reports describing paradoxical reactions during the treatment of *M. ulcerans*.

There is published histological evidence to support *M. ulcerans*-associated paradoxical reactions in patients being treated with antibiotics. Antibiotic therapy for *M. ulcerans* has been shown to lead to an apparent reversal of the immune-tolerant state of active *M. ulcerans* infection, with phagocytosis of mycobacteria and a rapid onset of local cellular immune responses.[1] These responses are characterised by the formation of epithelioid granuloma with multinucleated giant cells and large clusters of lymphocytes. It is likely that antibiotics facilitate this immune reaction by reducing the production of the immuno-inhibitory exotoxin mycolactone,[1] and also by liberating mycobacterial antigens from dead organisms.

The evidence for a paradoxical reaction in our cases included: (i) persisting *M. ulcerans* organisms in affected tissue after the initial surgery; (ii) an initial clinical improvement in the *M. ulcerans* lesions during antibiotic therapy, followed by significant clinical deterioration 1–2 months after antibiotic therapy was commenced; (iii) negative cultures from aspirates and excised tissue, suggesting that remaining mycobacteria were not alive as would be expected in a lesion that was worsening as a result of uncontrolled infection; (iv) the lack of mycobacteria seen on microscopy of the excised tissue, compared with active lesions that usually show large numbers of extracellular mycobacteria;[4] and (v) significant inflammatory reactions evident on histopathological examination of excised tissue, in contrast to what was observed in the initial active lesions, and consistent with recent descriptions of immune responses in successfully treated *M. ulcerans* infections.[1] Thus, we propose that rather than progression of the lesions because of failure of antibiotic treatment, these cases represent an adverse consequence of effective antibiotic treatment.

By applying lessons learnt from the management of immune-mediated reactions to infections with *Mycobacterium tuberculosis*,[2] the recognition of paradoxical reactions during treatment of *M. ulcerans* infections might significantly influence the management of these infections. First, rather than ceasing the antibiotic therapy

or changing the regimen, we advocate that therapy be continued, and that it not be changed unless there is evidence of antibiotic failure on histopathological examination or culture. Further, prolongation of the intended duration of antibiotic therapy is probably not required. Second, it may be possible that repeated needle aspiration for fluctuant lesions, as we described in Patient 1, may be effective in settling the lesion. This has been shown for lesions caused by *M. tuberculosis*,[11] and would avoid further surgery, which, in Patient 1, included costly and disfiguring skin grafts. Finally, adjunctive corticosteroid therapy may help to settle the lesion and obviate the need for, or reduce the extent of, surgical intervention.[12]

We feel that the descriptions of these cases are important, as they can alert clinicians to the possibility of paradoxical reactions occurring during antibiotic treatment for *M. ulcerans*. We recommend that when initial improvement on antibiotic treatment is followed by clinical deterioration of lesion, clinicians perform histopathological examination and mycobacterial culture of involved tissue to assess the possibility of a paradoxical reaction.

Competing interests

None identified.

Author details

Daniel P O'Brien, MB BS, FRACP, Infectious Diseases Specialist[1,2]
Michael E Robson, MB BS, FRCPA, Pathologist[3]
Peter P Callan, MB BS, FRACS, Plastic Surgeon[1]
Anthony H McDonald, MB BS, FRACS, Plastic Surgeon[1]
1 Barwon Health, Geelong, VIC.
2 Public Health Department, Médecins Sans Frontières, Amsterdam, Netherlands.
3 Pathcare, Geelong, VIC.
Correspondence: daniel.obrien@amsterdam.msf.org

References

1 Schutte D, Umboock A, Pluschke G. Phagocytosis of *Mycobacterium ulcerans* in the course of rifampicin and streptomycin chemotherapy in Buruli ulcer lesions. *Br J Dermatol* 2009; 160: 273-283.
2 Lipman M, Breen R. Immune reconstitution inflammatory syndrome in HIV. *Curr Opin Infect Dis* 2006; 19: 20-25.
3 Carvalho AC, De Iaco G, Saleri N, et al. Paradoxical reaction during tuberculosis treatment in HIV-seronegative patients. *Clin Infect Dis* 2006; 42: 893-895.
4 Guarner J, Bartlett J, Whitney EA, et al. Histopathologic features of *Mycobacterium ulcerans* infection. *Emerg Infect Dis* 2003; 9: 651-656.
5 Adusumilli S, Mve-Obiang A, Sparer T, et al. *Mycobacterium ulcerans* toxic macrolide, mycolactone modulates the host immune response and cellular location of *M. ulcerans* in vitro and in vivo. *Cell Microbiol* 2005; 7: 1295-1304.
6 Coutanceau E, Decalf J, Martino A, et al. Selective suppression of dendritic cell functions by *Mycobacterium ulcerans* toxin mycolactone. *J Exp Med* 2007; 204: 1395-1403.
7 O'Brien DP, Hughes AJ, Cheng AC, et al. Outcomes for *Mycobacterium ulcerans* infection with combined surgery and antibiotic therapy: findings from a south-eastern Australian case series. *Med J Aust* 2007; 186: 58-61.
8 Chauty A, Ardant MF, Adeye A, et al. Promising clinical efficacy of streptomycin–rifampin combination for treatment of Buruli ulcer (*Mycobacterium ulcerans* disease). *Antimicrob Agents Chemother* 2007; 51: 4029-4035.
9 World Health Organization. Provisional guidance on the role of specific antibiotics in the management of *Mycobacterium ulcerans* disease. Geneva: WHO, 2004.
10 Johnson PD, Hayman JA, Quek TY, et al. Consensus recommendations for the diagnosis, treatment and control of *Mycobacterium ulcerans* infection (Bairnsdale or Buruli ulcer) in Victoria, Australia. *Med J Aust* 2007; 186: 64-68.
11 Hawkey CR, Yap T, Pereira J, et al. Characterization and management of paradoxical upgrading reactions in HIV-uninfected patients with lymph node tuberculosis. *Clin Infect Dis* 2005; 40: 1368-1371.
12 Garcia Vidal C, Rodríguez Fernández S, Martínez Lacasa J, et al. Paradoxical response to antituberculous therapy in infliximab-treated patients with disseminated tuberculosis. *Clin Infect Dis* 2005; 40: 756-759.

(Received 5 May 2009, accepted 17 Aug 2009) ❑