Successful Treatment of *Balamuthia mandrillaris* Amoebic Infection with Extensive Neurological and Cutaneous Involvement

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Granulomatous amoebic encephalitis caused by *Balamuthia mandrillaris* is an uncommon infection for which there is no optimal therapy. We describe a young, female patient who presented with extensive cutaneous and neurological involvement and who recovered after receiving prolonged treatment with miltefosine, fluconazole, and albendazole.

*Balamuthia mandrillaris* is a free-living amoeba that has been recognized as an uncommon human pathogen since 1990 [1]. More than 150 cases of human encephalitis due to *B. mandrillaris* have been reported worldwide; most have been reported from Latin America and from the southwestern region of the United States [2]. The disease induced by this amoeba is characterized by involvement of the skin, especially around the central face following a chronic course, with subsequent extension to the central nervous system (CNS), where it causes multifocal granulomatous encephalitis, leading almost invariably to a fatal outcome [3–5]. No specific treatment is available to manage this lethal condition. A number of antimicrobials (alone and in combination) have been tried with unsuccessful results, including amphotericin B, azoles, paromomycin, albendazole, pentamidine isethionate, macrolides, metronidazole, sulfadiazine, and some others [5–7]. However, 6 patients (4 in the United States and 2 in Peru) who presented with extensive CNS involvement survived after receiving several antimicrobial regimens [4–6, 8]. We report, to our knowledge, the first successful treatment of a young patient with *Balamuthia* encephalitis with a combination regimen of miltefosine, fluconazole, and albendazole.

**Case report.** A 21-year-old woman presented in August 2006 with a long-term history of cutaneous lesions on her right knee. Four years before admission, she had noticed several papular, erythematous, and painless lesions that had appeared 2 weeks after a fall in front of her house. The lesions coalesced to form a violaceous and indurate plaque covering the entire right knee. Empirical therapy was started at another institution with topical antifungal creams that contained fluconazole and clotrimazole plus topical steroids, which were administered for almost 1 year, followed by a combination of oral fluconazole (150 mg per day for ~45 days) and subsequent addition of clarithromycin (1500 mg per day) and trimethoprim-sulfamethoxazole (TMP-SMX; 320 mg/1600 mg per day) for 8 months, without improvement.

The patient was born in Lima, Peru, and has resided there for her entire life. She denied travel or specific occupational exposure, including gardening and swimming in brackish, fresh, or sea water. She noticed enlargement of the lesion on the right knee and the appearance of 3 new papular lesions—to them around the plaque on the right knee, and the third on the left thigh—in February 2006. Both lesions subsequently evolved into violaceous plaques. She was first seen at our institution 4 months later, where a skin biopsy of the lesion on the right knee was performed (Figure 1A). Histopathologic examination revealed a dense inflammatory infiltrate of the dermis composed of lymphocytes, plasma cells, and ill-defined granulomas, with great number of multinucleated giant cells located inside and outside the granulomas. A microorganism with a nucleus that has a large, central karyosome and a vacuolated cytoplasm, compatible with an amoebic trophozoite, was observed (Figure 1B). In addition, *B. mandrillaris* was isolated from a skin sample cultured in an axenic culture prepared with monkey kidney cells [7]. Polymerase chain reaction of a skin sample also yielded positive results for *B. mandrillaris* (the amplification was performed using the primer mitochondrial 16S rRNA gene from *B. mandrillaris* as a target) [9]. A skin biopsy sample was sent to the Public Health Department of California (Sacramento) and to the Centers for Disease Control and Prevention (Atlanta, GA), where *B. mandrillaris* infection was confirmed by immunohistochemical staining in September 2006. A brain computed tomograph, without contrast enhancement, yielded normal findings at this time. Empirical therapy was started with itraconazole (200 mg per day), and albendazole (400 mg per day).
Figure 1. A, Cutaneous lesion on the right knee observed in February 2006 showing an indurated and violaceous plaque covering the entire knee with 2 papular lesions. B, Skin biopsy specimen showing a dense inflammatory infiltrate of the dermis with granulomas (hematoxylin and eosin stain). An amoebic trophozoite is observed, with a nucleus that has a large, central karyosome and vacuolated cytoplasm. C, Fluid-attenuated inversion recovery (FLAIR) magnetic resonance image (MRI) obtained 7 days after the onset of neurologic symptoms (June 2007) showing hypersignal in the left temporal lobe. D, Axial gadolinium-enhanced T1-weighted sequence (June 2007) showing a ring-enhancing lesion in the left temporal lobe. E, Follow-up of the left knee lesion 1 week after the patient had commenced treatment with miltefosine, albendazole, and fluconazole. The lesions abruptly changed, developing a scaly and crusty surface. F, Axial gadolinium-enhanced MRI obtained 5 months after the start of treatment, showing significant improvement on the neurological lesions without evidence of contrast enhancing. G, Gadolinium-enhanced MRI image 4 months after completion of treatment, showing the disappearance of the brain lesions. H, Follow-up of the healed left knee lesions (May 2008).

The enlargement of the lesions continued despite medical treatment, and the patient was hospitalized in October 2006 to receive amphotericin B (cumulative dose, 1.5 g), in addition to itraconazole and albendazole. The lesions improved initially, but reactivation of the lesion on the left thigh and the appearance of 2 new small papular lesions (one on the left breast and the other one on her back) were observed in February 2007. Biopsy of the lesion on the back was performed; the biopsy findings were the same as described above. A surgical resection of the new lesions was performed, and TMP-SMX (320 mg/1600 mg by day) was added to the treatment regimen, without improvement.

The patient was readmitted to the hospital in May 2007 because of reactivation of the lesion on the surgical wound on
<table>
<thead>
<tr>
<th>Patient</th>
<th>Age, years</th>
<th>Sex</th>
<th>Region</th>
<th>Type of lesion(s)</th>
<th>Treatment regimen</th>
<th>Outcome</th>
<th>Duration of follow-up</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>64</td>
<td>M</td>
<td>California</td>
<td>Skin: raised lesion on the right forearm; CNS: 4 ring-enhancing lesions (parietal and occipital lobes)</td>
<td>Flucytosine (8 g per day), fluconazole (400 mg per day), and sulfadiazine (6 g per day) for 5 years; clarithromycin (500 mg per day) for 2 years; and pentamidine isethionate (4 mg/kg per day) and trifluoperazine (20 mg per day) for 18 days</td>
<td>Performing all activities of daily living, with good communication skills</td>
<td>5 years</td>
<td>[5]</td>
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<tr>
<td>2</td>
<td>5</td>
<td>F</td>
<td>California</td>
<td>CNS: 2 ring-enhancing lesions (temporal and parietal lobes)</td>
<td>Flucytosine (110 mg/kg per day) and fluconazole (4 mg/kg per day) for 2.4 years; pentamidine isethionate (1 mg/kg per day) for 34 days; clarithromycin (14 mg/kg per day) for 2.4 years; and thioridazine (1 mg/kg per day) for 1.8 years</td>
<td>Typical school performance, without gross neurologic sequelae</td>
<td>2.4 years</td>
<td>[5]</td>
</tr>
<tr>
<td>3</td>
<td>72</td>
<td>F</td>
<td>New York</td>
<td>CNS: 2 ring-enhancing lesions (frontal and temporal lobes)</td>
<td>Fluconazole (400 mg per day), sulfadiazine (6 g per day), clarithromycin (1500 mg per day), and pentamidine isethionate (300 mg per day); duration of therapy is unknown</td>
<td>No significant neurological sequelae</td>
<td>6 months</td>
<td>[6]</td>
</tr>
<tr>
<td>4</td>
<td>35</td>
<td>M</td>
<td>California</td>
<td>CNS: focal enhancing lesions</td>
<td>NA</td>
<td>Alive and in good conditions</td>
<td>3 months</td>
<td>[8]</td>
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<tr>
<td>5</td>
<td>8</td>
<td>M</td>
<td>Piura, Peru</td>
<td>Skin: large plaque on the central face; CNS: multiple focal lesions (both hemispheres)</td>
<td>Albendazole (400 mg per day) and itraconazole (200 mg per day) for 14 months</td>
<td>Skin and CNS lesions healed; below-average to average school performance; mild left hemiparesis</td>
<td>3 years</td>
<td>[4]</td>
</tr>
<tr>
<td>6</td>
<td>10</td>
<td>F</td>
<td>Piura, Peru</td>
<td>Skin: large plaque on the central face; CNS: 2 ring-enhancing lesions (frontal and temporal lobe)</td>
<td>Albendazole (600 mg per day), itraconazole (100 mg per day), and TMP-SMX (320 mg/1600 mg per day) for 6 months; surgical resection</td>
<td>No neurological sequelae</td>
<td>18 months</td>
<td>[4]</td>
</tr>
<tr>
<td>7</td>
<td>21</td>
<td>F</td>
<td>Lima, Peru</td>
<td>Skin: multiple plaques (right knee, left thigh, breast, and back); CNS: 1 enhancing lesion (temporal lobe)</td>
<td>Itraconazole (200 mg per day) and albendazole (400 mg per day) for 10 months; amphotericin B (cumulative dose, 1.50 g) was added without improvement; TMP-SMX (320 mg/1600 mg per day) was added for 3 months; surgical resection of small lesions with posterior reaction; an additional course of amphotericin B (cumulative dose, 1.35 g); itraconazole (300 mg per day); TMP-SMX (640 mg/3200 mg per day) and albendazole (800 mg per day) was given for 45 days and did not yield improvement; clarithromycin (1500 mg per day) and artesunate (100 mg per day) were added for 2 weeks, without improvement, and 1 CNS lesion appeared in the brain MRI; therapy was changed to albendazole (800 mg per day) and fluconazole (450 mg per day) for 7.5 months, as well as miltefosine (150 mg per day for 12 days, followed by 100 mg per day for 7 months), at which time skin and CNS lesions healed</td>
<td>Skin and CNS lesions healed</td>
<td>30 months, including 12 without therapy and no recurrence</td>
<td>PR</td>
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**NOTE.** CNS, central nervous system (confirmed by MRI); MRI, magnetic resonance imaging; NA, not available; PR, present report; TMP-SMX, trimethoprim-sulfamethoxazole.
the left thigh. A second course of amphotericin B was started (cumulative dose, 1,350 g), in addition to an oral regimen of itraconazole (300 mg per day), albendazole (800 mg per day), and TMP-SMX (640 mg/3200 mg per day), for 1 month without improvement. In June 2007, clarithromycin (1500 mg per day) and oral artesunate (100 mg per day) were added to the regimen. At this time, the patient developed fever, headache, and childish behavior, with no other neurologic complaints. The results of serological tests for human immunodeficiency virus and human T lymphotropic virus–1 were negative. Magnetic resonance imaging of the brain performed 7 days after the beginning of neurologic symptoms demonstrated hyper-intensity in the left temporal lobe in the fluid-attenuated inversion recovery (FLAIR) sequence and a ring-enhancement lesion with gadolinium in the temporal lobe (Figure 1C and 1D). Treatment with amphotericin B, TMP-SMX, clarithromycin, and artesunate was discontinued; itraconazole was changed to fluconazole to optimize the penetration of the azole into the CNS; albendazole treatment was continued, and oral miltefosine (150 mg per day for 12 days and 100 mg per day thereafter) was added in mid-June 2007. The cutaneous lesions on the knee abruptly changed, developing a scaly, crusty surface (Figure 1E) 1 week after the patient started this regimen. The headache disappeared by the second week of this regimen, and fever waned by the fourth week. The skin lesion improved, becoming less scaly and flat, until it eventually disappeared. The patient did well, with regression of all neurologic symptoms. She was discharged home with the prescribed antibiotics in August 2007.

Significant improvement on the size of the neurological lesions was documented after 5 months of treatment (Figure 1F). The patient continued to take miltefosine, albendazole, and fluconazole for 7 months. The intracerebral lesions disappeared, as documented by magnetic resonance imaging performed in May 2008 (Figure 1G). She remains asymptomatic, all cutaneous lesions have healed (Figure 1H), and no recurrence has been recognized as of May 2010.

Discussion. Since the first report of a *Balamuthia mandrillaris* amoebic infection affecting a baboon in the San Diego Wild Animal Park in 1986 [1], >100 human cases have been reported worldwide, with only 6 survivors [4–8]. In 2003, Deetz et al [5] described 2 patients with extensive CNS involvement who responded to a prolonged treatment regimen (up to 5 years) that included fluocytosine, pentamidine isethionate, fluconazole, sulfadiazine, and a macrolide. A third patient with CNS involvement who responded to a regimen similar to the one described above, but without fluocytosine, was described by Jung et al [6]. A fourth patient from the California Encephalitis Project (CEP) was reported alive and in good health condition three months after diagnosis, details about his treatment were not provided however [8]. Two additional Peruvian patients survived long-term therapy (6 months) with albendazole and itraconazole, and another patient, who demonstrated cutaneous involvement alone, was treated with surgical resection plus a combination of itraconazole, albendazole and amphotericin B (cumulative dose, 2 g) [3, 4]. A description of the 7 patients who survived *Balamuthia* infection of the CNS, including the patient in our study, is presented in Table 1.

Very few drugs have shown in vitro amebicidal activity against *B. mandrillaris*, including miltefosine [10], propamidine, and ganciclovir S [7]. The amebicidal activity was demonstrated by drug efficacy test using axenic cultures inoculated in tissue culture flasks with growing concentrations of antimicrobials. Most of the available drugs are amebistatic: phenothiazine compounds, pentamidine isethionate, macrolides, azoles, TMP-SMX, and amphotericin B [7]. In addition, *B. mandrillaris* escapes the effect of antimicrobials by encysting in tissues, establishing chronic infections that may reactivate later [11]. Use of amebicidal drugs for treating this condition may not only kill trophozoites in active lesions, but may also prevent the further dissemination of the infection to the CNS and other organs, which has been invariably observed when amebistatic drugs are used.

Miltefosine is an alkylphosphocholine compound originally developed as an anticancer drug, which is now established as an effective anti-leishmanial therapy. It acts on key enzymes involved in phospholipid and sterol biosynthesis, suggesting that the cell membrane is its main target [12]. In vitro, it stimulates T cells and macrophages to secrete activating cytokines, including interferon-γ, and induces production of microbicidal reactive nitrogen and oxygen intermediates, causing cell death by an apoptosis-like effect [13]. A recent report showed good in vitro activity of miltefosine against *Balamuthia, Acanthamoeba*, and *Naegleria* species [10]. Miltefosine achieves a high concentration of the active drug in the brain tissue as a result of excellent passage through the brain-blood barrier. In addition, miltefosine carries an acceptable safety profile [10]. This is a potentially critical property of any drug to be successful in treating CNS involvement by free-living amebas.

Although the prognosis of amebic encephalitis caused by *B. mandrillaris* is still ominous, it may not be invariably fatal. This report on the successful use of a combination regimen that includes 1 amebicidal drug (miltefosine) along with 2 amebistatic drugs capable of crossing the brain-blood barrier (fluconazole and albendazole) provides hope for attaining clinical cure for an otherwise lethal condition. More clinical experience is needed before miltefosine can be recommended as the definitive first-line treatment for amebic encephalitis.

Acknowledgments

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References