Eradication of AIDS-Related Disseminated *Mycobacterium avium* Complex Infection after 12 Months of Antimycobacterial Therapy Combined with Highly Active Antiretroviral Therapy

Judith A. Aberg, David M. Yajko, and Mark A. Jacobson

To determine if microbiologic cure of AIDS-related disseminated *Mycobacterium avium* complex (MAC) is possible in patients receiving highly active antiretroviral therapy (HAART), 4 patients with a history of disseminated MAC received ≥12 months of macrolide-based antimycobacterial therapy. All were asymptomatic and had absolute CD4 cell count >100/μL (range, 137–301) and <10,000 copies/mL of human immunodeficiency virus RNA (range, <500–1250). A bone marrow aspirate and peripheral blood were obtained for mycobacterial culture. Follow-up blood cultures were obtained routinely at 4 weeks and every 8 weeks thereafter. All 4 patients had negative bone marrow and blood cultures and then discontinued antimycobacterial therapy. All patients’ subsequent cultures remain sterile and all are clinically asymptomatic (range, 8–13 months follow-up). It appears that disseminated MAC infection can be cured by prolonged antimycobacterial therapy in some persons who experience sustained CD4 lymphocyte increases while receiving HAART.

Prior to the introduction of highly active antiretroviral therapy (HAART), disseminated infection with *Mycobacterium avium* complex (MAC) was one of the most common life-threatening opportunistic infections affecting patients with AIDS and occurred almost exclusively in patients with <50 CD4 cells/μL. Prognosis was poor with a median survival of only 8–9 months from the diagnosis of disseminated MAC, even with optimal antimycobacterial treatment [1–4]. Despite the widespread use of a multidrug macrolide-based regimen, relapses were common [5]. Standard of care was to continue treatment lifelong, since microbiologic cure was not reported [6, 7]. However, we have observed patients who developed disseminated MAC with <50 CD4 cells/μL, who after subsequent HAART have had sustained immunologic responses with persistent absolute CD4 cells >100/μL. These patients have received antimycobacterial therapy for disseminated MAC for >1 year and are now asymptomatic and medically stable. We hypothesized that if HAART reconstituted MAC-specific immunity, a 12-month course of antimycobacterial treatment would be curative. Because bone marrow can harbor infection in the presence of sterile blood cultures [8], we confirmed absence of MAC in both blood and bone marrow before discontinuing MAC treatment.

Methods

Primary care providers at San Francisco General Hospital identified 4 patients with a history of disseminated MAC who had received macrolide-based therapy for ≥12 months, were asymptomatic, and had ≥1 absolute CD4 cell count of >100 cells/μL and plasma human immunodeficiency virus (HIV) RNA of <10,000 copies/mL after HAART. Two peripheral blood cultures and a bone marrow aspirate for culture were obtained. Antimycobacterial therapy was discontinued after cultures revealed no growth for 6 weeks. Follow-up blood cultures were obtained at 4 weeks and every 8 weeks thereafter. Blood cultures and bone marrow culture were to be obtained if any clinical signs or symptoms suggested recurrence of MAC infection.

Blood cultures were processed via the BACTEC 12B system. Heparinized blood was obtained and centrifuged for 10 min at 3000 g. In total, 0.5 mL of the buffy coat was pipetted and injected into a BACTEC 12B vial.

Bone marrow aspirates were centrifuged, and an acid-fast bacilli smear was made from the sediment. The sediment was then resuspended, and 0.5 mL was transferred to a BACTEC 12B vial. In addition, two drops of sediment was inoculated onto 7H11 agar.
Case Reports

Information regarding MAC diagnosis, numbers of CD4 cells, and HIV load is summarized in table 1.

Patient 2, a 42-year-old African-American man with a history of *Cytomegalovirus* retinitis, was diagnosed with MAC bacteremia in May 1995. He reported a 1-month history of fever and weight loss. He had 4 CD4 cells/μL. Peripheral blood cultures from June 1995 grew MAC. He enrolled in ACTG 223 protocol and was treated with clarithromycin, ethambutol, and rifabutin. Per ACTG 223 protocol, he had blood cultures obtained at study entry, at weeks 4, 6, and 8, and every 4 weeks for 48 weeks in total. He had persistent positive quantitative blood cultures for MAC in May, June, and July 1995; however, the cultures became sterile in August 1995, and with MAC therapy his symptoms resolved. Rifabutin was discontinued in November 1996. At that time, he was started on a regimen of zidovudine, lamivudine, and indinavir when his virus load was 119,800 copies/mL. In January 1997, he had 218 CD4 cells/μL and a virus load of 1250 copies/mL. After negative blood and bone marrow cultures were obtained, antimycobacterial therapy was withdrawn in March 1997. At that time, the patient’s CD4 cells totaled 301/μL and his virus load was 1250 copies/mL. As of March 1998, the patient remained asymptomatic, and all subsequent blood cultures have been negative. His CD4 cell count is >300/μL and his virus load remains <15,000 copies/mL.

Patient 3, a 28-year-old Caucasian woman, was diagnosed with MAC bacteremia in July 1996. She reported fever and abdominal pain and was anemic. She had 4 CD4 cells/μL. An abdominal computed tomography (CT) scan revealed extensive lymphadenopathy with involvement of the gastrohepatic, porta hepatis, paraceliac, bilateral renal hilar, mesenteric, obturator, and common iliac nodal chains. Two blood cultures drawn 9 days apart grew MAC. She was treated with clarithromycin and ethambutol and began zidovudine and lamivudine therapy at the time of MAC diagnosis. Her symptoms resolved, and blood cultures became sterile after several weeks of therapy. Indinavir was added to her regimen in October 1996. In November 1996, she had 93 CD4 cells/μL and a virus load of <500 copies/mL. A repeat abdominal CT scan in December 1996 showed decreased lymphadenopathy. In January 1997, azithromycin replaced clarithromycin in her treatment regimen and in February 1997, nelfinavir replaced indinavir. At that time, she had 157 CD4 cells/μL and her virus load remained undetectable. In May 1997, she had 202 CD4 cells/μL and her virus load was 516 copies/mL. In June 1997, her virus load was <500 copies/mL. After obtaining negative blood and bone marrow cultures, antimycobacterial therapy was withdrawn in July 1997. On 30 July 1997, all antiretrovirals were changed to her current regimen of didanosine, stavudine, and combination saquinavir-ritonavir. In January 1998, her CD4 cell count was 389 cells/μL and her virus load was 5384 copies/mL. As of March 1998, the patient remained asymptomatic; all subsequent blood cultures have been negative.

Patient 4, a 26-year-old Vietnamese man, was diagnosed with MAC bacteremia in March 1996. At that time, he reported fever, weight loss and cough, and shortness of breath. He had 4 CD4 cells/μL. Peripheral blood cultures, bronchial alveolar lavage, and a subcarinal lymph node biopsy grew MAC. He was treated with clarithromycin and ethambutol; zidovudine and lamivudine therapy were initiated at the time of MAC diagnosis. His symptoms resolved and blood cultures became sterile after several weeks of therapy. Ritonavir was added to his regimen in August 1996. In September 1996, he had 103

May 1997. A repeat CD4 cell count was 137 cells/μL; again the virus load remained undetectable. As of March 1998, the patient remained asymptomatic; all subsequent blood cultures have been negative. His most recent CD4 cell count was 200 cells/μL. His virus load remained undetectable.

Patient 4, a 26-year-old Vietnamese man, was diagnosed with MAC bacteremia in March 1996. At that time, he reported fever, weight loss and cough, and shortness of breath. He had 4 CD4 cells/μL. Peripheral blood cultures, bronchial alveolar lavage, and a subcarinal lymph node biopsy grew MAC. He was treated with clarithromycin and ethambutol; zidovudine and lamivudine therapy were initiated at the time of MAC diagnosis. His symptoms resolved and blood cultures became sterile after several weeks of therapy. Ritonavir was added to his regimen in August 1996. In September 1996, he had 103

### Table 1. Patient CD4 cells at time of *Mycobacterium avium* complex (MAC) diagnosis and subsequent CD4 cell counts and human immunodeficiency virus (HIV) loads at time antimycobacterial therapy was discontinued.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Date MAC diagnosed</th>
<th>No. of CD4 cells at MAC diagnosis</th>
<th>Date therapy discontinued</th>
<th>No. of CD4 cells when MAC therapy ended</th>
<th>HIV load when MAC therapy ended</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5/95</td>
<td>5</td>
<td>3/97</td>
<td>301</td>
<td>1250</td>
</tr>
<tr>
<td>2</td>
<td>6/95</td>
<td>4</td>
<td>5/97</td>
<td>137</td>
<td>&lt;500</td>
</tr>
<tr>
<td>3</td>
<td>7/96</td>
<td>4</td>
<td>7/97</td>
<td>202</td>
<td>&lt;500</td>
</tr>
<tr>
<td>4</td>
<td>3/96</td>
<td>4</td>
<td>8/97</td>
<td>220</td>
<td>&lt;500</td>
</tr>
</tbody>
</table>

NOTE: Cells/μL; HIV virus load, copies/mL.
CD4 cells/µL and a virus load of <500 copies/mL. After negative blood and bone marrow cultures were obtained, antimycobacterial therapy was withdrawn in August 1997 when he had 220 CD4 cells/µL and a virus load of <500 copies/mL. Since October 1997, he has had >250 CD4 cells/µL and his virus load has remained undetectable. As of March 1998, he remained asymptomatic and all subsequent blood cultures have been negative.

Discussion

To our knowledge, this is the first report in which antmycobacterial therapy for disseminated MAC has been successfully discontinued in patients with AIDS after presumed immune reconstitution following HAART. There were no reports of microbiologic cure in the pre-HAART era. Administration of chronic suppressive antimycobacterial therapy was lifelong and relapse was associated with discontinuation of antimycobacterials. Although median survival with optimal antimycobacterial treatment in the pre-HAART era was <1 year, our 4 patients were asymptomatic after 1 year of combined antimycobacterial therapy and potent antiretroviral therapy, had sterile blood and bone marrow cultures, and had no clinical or microbiologic evidence of MAC relapse 8–13 months after discontinuing antimycobacterial therapy. These observations suggest that AIDS-related disseminated MAC can be cured in patients capable of immunologically improving with HAART.

The mechanisms of pathogenesis of disseminated MAC are still not completely understood. The role of colonization of the gastrointestinal and respiratory tracts, the timing and sites of invasion, the significance of multiple colonizing strains, and the interplay of various immune defenses remain unknown. One hypothesis is that MAC disease starts with colonization of the gastrointestinal or respiratory tract and then establishes a localized infection with intermittent MAC bacteremia leading to seeding of other organs. This is followed by an unknown period of proliferation in these extravascular sites until the increasing organism burden results in sustained symptoms and “spillover” sustained bacteremia. This model is supported by studies from autopsies of HIV-infected patients with MAC bacteremia, which report MAC concentrations in multiple organs to be many times higher than concentrations in the blood. Even in patients with relatively few disease-associated symptoms, the levels of MAC cultured from bone marrow samples are typically several logs higher than levels in blood [8]. This model suggests that most persons with symptomatic MAC bacteremia will have widespread organ involvement and that they may not have consistently detectable bacteremia until the spillover from tissue occurs.

Kemper et al. [9] reported 9 patients in whom MAC bacteremia became undetectable before initiation of MAC therapy. Subsequently, 6 of the 9 patients became mycobacteremic again. More recently, Hadad et al. [10] reported a patient who grew a single colony from 1 peripheral blood culture. After HAART initiation, symptoms resolved completely and no further cultures were positive for MAC. This supports the hypothesis that a transient bacteremia precedes MAC dissemination and that the immunologic consequences following HAART may be able to prevent dissemination.

Our patients differ from those with transient bacteremia. As described above, 3 of the 4 patients had sustained bacteremia and the 4th had a positive culture from two other body sites, including a sterile tissue source.

Race et al. [11] reported 5 patients who developed a focal mycobacterial lymphadenitis within 1–3 weeks of starting antiretroviral therapy with indinavir. They hypothesized that an intense inflammatory reaction resulted from a significant expansion of a subset of CD4 cells with MAC-specific immunity responding to a heavy mycobacterial tissue burden (that existed as subclinical infection before initiation of antiretroviral therapy). We did not note any atypical presentations in the 2 patients who began MAC and antiretroviral therapy concomitantly.

Host immune mechanisms are responsible for the absence of invasive MAC disease in normal hosts, and general or MAC-specific impairments in immunity lead to the increasing incidence of disseminated MAC as HIV disease progresses. Although the precise interactions are unknown, the key host defense mechanisms against mycobacterial infection include CD4+ and CD8+ lymphocytes, and more proximally, macrophages, NK cells, and various cytokines, many of which are CD4 cell dependent. HIV replication is characterized by ongoing viral replication that leads to progressive depletion of CD4 cells with preferential loss of naive cells. Some studies report that increases in CD4 cells with HAART represent expansion of the remaining elements of the repertoire rather than addition of new or reacquisition of the lost cells [12, 13]. However, other studies indicate that with persistent suppression of HIV replication, increases in both memory and naive CD4 cells can occur [14, 15]. Therefore, it is plausible that HAART interruption of HIV replication could permit CD4 repopulation to reconstitute MAC protective immunity.

Among the cytokines that appear critical for MAC protection are interferon (IFN)-γ, interleukin (IL)-2, granulocyte colony-stimulating factor (CSF), granulocyte-macrophage CSF, tumor necrosis factor-α, and IL-12 [16]. The function or synthesis of IFN-γ, IL-2, and IL-12 are particularly CD4 cell dependent. Production of IFN-γ declines significantly in vitro with only modest reductions in CD4+ cells [17]. Immunity to M. tuberculosis appears to be especially dependent on the production of IFN-γ by CD4+ cells; therefore, this finding may relate to the increased incidence of tuberculosis in HIV-infected persons with early HIV disease and relatively high numbers of CD4+ cells. In contrast, disseminated MAC infection usually does not occur in AIDS patients until CD4+ cells fall to <50/mm³, which suggests that additional CD4+-dependent factors may contrib-
ute to natural defense against MAC. The relationship between the cell types and cytokine profiles identified in the blood are not completely understood. Several ACTG studies are in various phases of development to further describe the MAC protective immunity that occurs with immune reconstitution.

Future studies are needed to further characterize the immunopathogenesis of disseminated MAC so that persons at highest risk for MAC infection can be identified. It would then be possible to target prophylaxis or preemptive therapy for that population and to discontinue that treatment if HAART restores immune responses associated with MAC protection. In addition, better understanding of immune mechanisms would provide data for the design and development of immune-based therapies and assist in predicting which patients with disseminated MAC can be expected to be cured of their infection after a course of antimycobacterial treatment.

References