Unmasked Tuberculosis and Tuberculosis Immune Reconstitution Inflammatory Disease: A Disease Spectrum after Initiation of Antiretroviral Therapy

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Antiretroviral therapy (ART) has beneficial effects on mortality and lowers the incidence of diseases caused by opportunistic infections, such as tuberculosis (TB). Although ART has sustained long-term benefits, the risk of TB is high during the first 3 months after ART initiation. Among cases of ART-associated TB, we define “unmasked TB” as that which occurs in patients with reactivation disease who develop clinically recognizable TB after ART with the restoration of previously acquired TB antigen–specific functional immune responses. TB cases with clinical evidence of an inflammatory syndrome are a subset of these unmasked cases, which we define as “unmasked TB–immune reconstitution inflammatory syndrome.” With more widespread use of ART in areas with a high prevalence of TB, unmasked TB will likely become more common. TB diagnostics with improved sensitivity and specificity are urgently needed to detect subclinical TB before it is unmasked.

The global increase in the number of tuberculosis (TB) cases has been driven by the HIV/AIDS pandemic, particularly in sub-Saharan Africa [1]. Antiretroviral therapy (ART) improves HIV morbidity and mortality and also decreases the risk of developing TB by 70%–90% [2–4]. Although the risk of TB in patients receiving ART becomes lower in the long term because of progressive immune reconstitution, the risk is highest during the first few months after the initiation of ART [5–9]. The development of TB after ART initiation, or “ART-associated TB,” has become an increasingly important problem [10]. Among ART-associated TB cases, we define “unmasked TB” as that which occurs in the subset who develop clinically recognizable TB after the initiation of ART because of the restoration of TB antigen–specific functional immune responses. Only a proportion of individuals with these unmasked TB (reactivation) cases will develop rapid, often destructive necrotic inflammation that is consistent with immune reconstitution inflammatory syndrome (IRIS). We propose to call these cases “unmasked TB-IRIS” (figure 1A and 1B). Therefore, unmasked TB excludes patients whose TB is primary and is only temporally associated with ART (i.e., these patients do not have rapid recall of TB antigen–specific memory immune responses). In this group of patients with primary TB, it would be unlikely that TB-IRIS would develop soon after the initiation of ART.

In this case report and review of the literature, we present the disease spectrum of TB after the initiation of ART. We also report a case of unmasked TB-IRIS: pulmonary and lymphatic TB that became clinically evident in a patient with advanced AIDS only after the initiation of ART and virologic suppression. The clinical presentation of TB was consistent with TB-IRIS, and the disease was unmasked because it was not diagnosed before ART despite an extensive evaluation. Moreover, we
present the results of a review of the literature for incidence rates of TB occurring within the first 3 months after ART initiation, which identified reports of ART-associated TB and of the smaller subset of unmasked TB-IRIS cases. Finally, we propose a novel model of unmasked TB and TB-IRIS in which the CD4 T cell count at the time of ART initiation is critical; as CD4 T cell counts decline, patients coinfected with HIV and *Mycobacterium tuberculosis* are more likely to reactivate latent TB infection and have increased bacillary burden because of ongoing replication. Early identification and treatment of TB before ART initiation, as well as prompt recognition and treatment of unmasked TB and unmasked TB-IRIS, could impact the TB pandemic.

**CASE REPORT**

A 25-year-old African American man presented with chronic watery diarrhea, subjective fevers, chills, and loss of 30 pounds of weight over 1 month. He had no respiratory symptoms. He was known to be HIV infected and had a peripheral blood CD4 T cell count of 32 cells/µL and an HIV-1 RNA level of 298,000 copies/mL. He was ART naïve. There was no measurable induration in his Mantoux skin test. Results of stool and blood cultures and of fungal antigen testing were negative for bacterial, mycobacterial, and fungal pathogens. A chest radiograph was notable for a linear soft-tissue opacity at the right tracheal border, reported by the radiologist as being compatible with vasculature (figure 2A). He had no cough and was unable to produce an induced sputum specimen. A gastrostomy tube was placed 1 month after admission, because the patient had organic brain syndrome from prior head trauma and HIV-related dementia and was unable to take medications orally. Placement of the gastrostomy tube was complicated by a posterior stomach wall perforation, and emergent laparotomy was required to repair the tear; a gastrojejunostomy tube was placed. The patient received a postoperative course of piperacillin-tazobactam for an acute abdomen and a right lower lobe infiltrate (presumed aspiration pneumonia), with rapid radiographic resolution on β-lactam therapy (which has only weak antimycobacterial activity). ART (tenofovir, emtricitabine, and ritonavir-boosted lopinavir) was initiated 2 weeks after this admission.

Twenty days after commencing ART, the patient presented with a fever of 104°F and cough. At this time, a repeat Mantoux test produced a positive result (24 mm of induration). A chest radiograph (figure 2B) and computerized tomography showed extensive consolidation of the right upper lobe with areas of cavitation and a heterogeneous mass extending 9 cm from the right midneck down the medial right hemithorax. An induced sputum sample was negative for acid-fast bacilli (AFB) on auramine smear. Bronchoalveolar lavage was also AFB smear negative. However, Kinyoun stain of a lymph node transbronchial needle aspirate obtained at bronchoscopy was positive for AFB. All sputa and bronchoalveolar lavage culture results were subsequently positive for *M. tuberculosis* with in vitro sensitivity to all first-line anti-TB drugs. At the time of TB-IRIS presentation 3 weeks after the initiation of ART, the patient’s HIV-1 RNA level had decreased to 1982 copies/mL.

Informed consent was obtained from the patient presented to publish the details of his medical presentation and management.

**METHODS FOR AND RESULTS OF LITERATURE SEARCH**

We performed a computer-based search of published literature (MEDLINE, National Library of Medicine, Bethesda, Maryland) using the following keywords: “antiretroviral,” “ART,” “TB,” “mortality,” “immune reconstitution,” and “immune restoration.” Data were abstracted from these articles as well as from the references in these articles. The search identified 21 studies in which TB cases that occurred after ART initiation were presented. Seven of these studies reported the number of patients who presented during the first 3 months after ART initiation, and 3 additional studies reported TB incidence rates during the first 3 months (table 1).

**REVIEW AND DISCUSSION**

**Cases of TB within 3 months after ART initiation.** Although many cases of *Mycobacterium avium* disease unmasking have been reported after the initiation of ART [15], there have been substantially fewer reports of unmasked disease due to *M. tuberculosis*. Most case reports have been from developed countries with low rates of TB [2, 5, 16, 17], although a few reports from resource-limited settings have also been published (table 2). In the majority of these studies (as in our case), the patients described had the inflammatory features of IRIS and, therefore, were classified as having cases of unmasked TB-IRIS. This represents only a small (but clinically impressive) subset of all cases of unmasked TB. We focused a larger search on cases that occurred within the first 3 months after ART initiation because the likelihood of reactivation TB was higher than that of newly acquired primary disease, although the latter likely also occurs. Interestingly, many of the patients had constitutional symptoms before ART initiation. Patients with clinically active TB before

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**Figure 1.** Unmasked tuberculosis (TB).

**Figure 2.** Chest radiograph from a case patient 1 month before antiretroviral therapy (ART) was initiated (A) and at the time of presentation 20 days after ART was initiated (B).
ART initiation are screened on the basis of pulmonary symptoms. Data from a TB-endemic area in South Africa showed that 5% of those screened (by culture) were positive and had a lack of clinical symptoms despite transmissible disease [20]. Furthermore, screening by smear microscopy (and not culture) will miss up to 50% of HIV-positive patients with active TB [21]. Finally, even in settings in which chest radiography can be performed, it is well recognized that the appearance of chest radiographs can vary according to the degree of immunosuppression in HIV-infected patients with confirmed TB (including normal chest radiographic findings) [22]. In a TB vaccine study in Tanzania, 11% of asymptomatic HIV-infected volunteers (CD4 T cell counts >200 cells/mL) had subclinical TB with normal chest radiographic findings but positive sputum culture results [23]. This highlights the existence of a reservoir of patients with unrecognized TB who may develop unmasked disease after immune reconstitution.

Reports from resource-limited settings as well as from the developed world (Table 1) have uniformly shown more cases of TB during the first 3 months after ART initiation than during later time periods [7, 8]. Similarly, in a large analysis of data from multiple observational databases, both resource-limited and industrialized countries showed the highest incidence rates during

<table>
<thead>
<tr>
<th>Study, location</th>
<th>Years of study</th>
<th>Duration, monthsa</th>
<th>Patients receiving ART, total</th>
<th>Patients presenting with TB &lt;3 months after ART</th>
<th>CD4 T cell count before ARTc</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bonnet et al. [11] Combined</td>
<td>2001–2004</td>
<td>3151</td>
<td>320</td>
<td>192 (60%)</td>
<td>131</td>
</tr>
<tr>
<td>Cambodia</td>
<td>. . .</td>
<td>3.7–12.8</td>
<td>717</td>
<td>100</td>
<td>63 (63%)</td>
</tr>
<tr>
<td>Thailand</td>
<td>. . .</td>
<td>1.3–8.3</td>
<td>500</td>
<td>57</td>
<td>30 (53%)</td>
</tr>
<tr>
<td>Kenya</td>
<td>. . .</td>
<td>1.4–8.0</td>
<td>654</td>
<td>73</td>
<td>41 (56%)</td>
</tr>
<tr>
<td>Malawi</td>
<td>. . .</td>
<td>3.2–15.4</td>
<td>1064</td>
<td>80</td>
<td>50 (63%)</td>
</tr>
<tr>
<td>Cameroon</td>
<td>. . .</td>
<td>3.2–19</td>
<td>216</td>
<td>10</td>
<td>8 (80%)</td>
</tr>
<tr>
<td>Breen et al. [5], England</td>
<td>1997–2004</td>
<td>. . .</td>
<td>19</td>
<td>11 (58%)</td>
<td>3</td>
</tr>
<tr>
<td>Park et al. [12], South Korea</td>
<td>1998–2005</td>
<td>31</td>
<td>482</td>
<td>27</td>
<td>5/9 (56%)d</td>
</tr>
<tr>
<td>Seyler et al. [6], Cote d’Ivoire</td>
<td>1996–2003</td>
<td>26</td>
<td>129</td>
<td>12</td>
<td>3 (25%) [4.8/100]e</td>
</tr>
<tr>
<td>Moore et al. [13], Uganda</td>
<td>2003–2005</td>
<td>16.8</td>
<td>1044</td>
<td>53</td>
<td>23 (43%) [3.9/100]f</td>
</tr>
<tr>
<td>Lawn et al. [9], South Africa</td>
<td>2002–2005</td>
<td>10.4</td>
<td>756</td>
<td>105g</td>
<td>22/100 [10.5/100]</td>
</tr>
<tr>
<td>Moh et al. [14], Cote d’Ivoire</td>
<td>2002–2004</td>
<td>8</td>
<td>792</td>
<td>25</td>
<td>5.6/100 [1.1/100]</td>
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<tr>
<td>ART-CC [8], high-income countries</td>
<td>Various–2006</td>
<td>. . .</td>
<td>22,217</td>
<td>205</td>
<td>1.7/100 [1.0/100]</td>
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<tr>
<td>ART-LINC [8], low-income countries</td>
<td>Various–2006</td>
<td>. . .</td>
<td>4810</td>
<td>258</td>
<td>10.7/100 [7.4/100]</td>
</tr>
<tr>
<td>Girardi et al. [7], Europe and North America</td>
<td>1996–2003</td>
<td>33.6</td>
<td>17,142</td>
<td>173</td>
<td>1.31/100 [0.47/100]</td>
</tr>
</tbody>
</table>

NOTE. Data are no. of patients, unless otherwise specified. ART-CC, ART Cohort Collaboration; ART-LINC, ART in Low-Income Countries Collaboration; EPTB, extrapulmonary TB; PTB, pulmonary TB.

Table 2. Clinical characteristics of patients with exacerbation of subclinical tuberculosis (TB) after antiretroviral therapy (ART) immune reconstitution.

The table is available in its entirety in the online edition of the Journal of Infectious Diseases.
the first 3 months [8]. Although increased clinical vigilance after the initiation of ART contributes to increased ascertainment of TB cases and highlights the possibility that high incidence rates during the first 3 months result from cases being unrecognized before ART initiation, there is also evidence for unmasked TB in developed countries, in which definitive diagnostic methods can be used. Therefore, we believe that unmasked TB contributes to higher early incidence rates in areas of endemnicity and points to a potentially larger problem as multiple large-scale programs that bring ART to the developing world are expanded [24].

**Effect of ART on the immune response to mycobacterial antigens and burden: a model.** Even with more-vigorous screening, TB incidence rates during the first 3 months after ART initiation are likely to be higher than during later time periods. The amount of mycobacterial antigen varies with the level of immunosuppression, such that patients with lower CD4 T cell counts have higher mycobacterial burdens (as suggested by the positive correlation between increasing immunosuppression and increased risk for disseminated TB with high bacillary burden) (figure 3) [25]. During immune reconstitution, the response to the mycobacterial antigen burden is potentiated as the functional cell-mediated immune response recovers with viral load suppression and CD4 T cell recovery [26]. When ART is initiated at higher CD4 T cell counts (time A), the potentiation of the immune response to *M. tuberculosis* antigens leads to further control of relatively low bacillary load and suppression of clinically evident and diagnosable TB. At lower CD4 T cell counts (time B), the number of unmasked (clinically evident) TB cases increases because the relative change in the immune response is greater per unit of mycobacterial antigen due to immune reconstitution (figure 3, shaded hatched area). In this model, clinically latent disease reactivates; in others, subclinical disease becomes clinically evident [5]. The majority of cases are identified during the first 3 months after ART in observational cohorts with the lowest CD4 T cell counts at ART initiation. Because of widespread access to ART, most patients who initiate ART in resource-limited settings do so while profoundly immunosuppressed (table 1). High rates of early mortality that correlate with CD4 T cell count nadir suggest that early mortality [27]
may be due to unrecognized or unmasked TB and/or other diseases caused by prevalent opportunistic infections.

There is some evidence from the rabbit model of TB that bacillary replication also increases after immune reconstitution, resulting in a higher mycobacterial burden to which an immune response is mounted [28]. After the initiation of ART, if immune reconstitution leads to a concomitant rise in the mycobacterial antigen burden, the number of unmasked TB cases would be even higher (figure 3, cross-hatched area). Furthermore, localization of increased amounts of mycobacterial antigen at particular sites (e.g., lungs and lymph nodes) can lead to localized exaggerated responses in unmasked TB-IRIS.

It is important to note that this model applies only to patients who have latent TB before the development of HIV-induced immunosuppression. We postulate that, in these patients, previously established memory immune responses are up-regulated during immune reconstitution. Therefore, immunosuppressed HIV-infected patients who develop primary TB are less likely to augment M. tuberculosis–specific immune responses soon after ART initiation because they do not yet have established memory responses (figure 1B). Conversely, patients with exogenous re-infection may have unmasked TB-IRIS soon after ART initiation because they could have existing M. tuberculosis–specific memory responses that could be potentiated with re-infection. Finally, the curve of the mycobacterial antigen increase with advancing immunosuppression varies according to the underlying genetic background of the host. There will be variation in the crossover point between falling CD4 T cell counts and increasing mycobacterial burden (figure 4). The T cell count at which unmasking can occur is, therefore, not a uniform threshold and will vary from person to person.

**TB-IRIS.** The importance of the host immune response in the pathogenesis of pulmonary TB and the formation of cavitary disease is well known [29]. In our case report, the patient presented with nonspecific constitutional symptoms that, after immune reconstitution, progressed to typical pulmonary cavitary disease that was diagnosed on the basis of sputum and bronchoalveolar lavage cultures positive for M. tuberculosis. In retrospect, the opacity on the right tracheal border was probably mediastinal lymphadenopathy harboring mycobacteria. The necrotic lymphadenopathy and cavitary disease likely occurred as a result of rapid virologic suppression and reconstituted CD4 T cell responses, because the patient was able to mount a vigorous delayed-type hypersensitivity response that had previously been negative. This restoration of mycobacterial antigen–specific immune responses is consistent with IRIS, which has been reported in 7%–36% of patients who begin ART within 2 months of antituberculous therapy [30–33]. The term “paradoxical worsening” has also been used to describe patients with known TB who deteriorate clinically (usually after initial improvement) after the commencement of antituberculous therapy [34, 35]. TB-IRIS describes HIV-infected patients who manifest clinical symptoms and signs consistent with an infectious or inflammatory condition after ART initiation and evidence of immune reconstitution [15, 36]. Although we have described a patient from a setting with a low incidence of TB, such cases occur in settings of both low and high TB incidence.

Not all of these patients with reactivated, clinically evident TB will develop IRIS. The risk for IRIS increases with the degree of immunosuppression at the time of ART initiation (figure 3). Patients with the lowest CD4 T cell counts are at the highest risk of developing IRIS. Other risk factors for IRIS include the rapidity of the viral load decline [37], bacillary/antigen load (disseminated disease), and genetic predisposition [38]. This risk must be balanced against the benefit of initiating ART early in patients with advanced immunosuppression [39]. In a decision analysis pertaining to patients coinfected with HIV and TB bacilli, there was a reduction in the risk of opportunistic infections and death with early (i.e., within 2 months of initiating TB therapy) initiation of ART [40]. The risks of early ART initiation exceeded the benefits only when IRIS-related mortality rates were high.

Evidence for the importance of bacillary antigen load in the development of IRIS is based on the increased incidence of IRIS in patients with disseminated disease and the shorter time interval between ART initiation and IRIS in patients with unmasking, compared with that in patients already receiving specific treatment for the opportunistic infection [37]. With TB, dead bacillary by-products and antigens can also trigger IRIS and cause it to persist despite TB treatment. Finally, in the rabbit model of TB-IRIS, rabbits infected with higher aerosol doses—and, therefore, with greater numbers of bacteria at the time of immuno-
suppression and subsequently at the time of immune reconstitution—were more likely to develop IRIS than rabbits infected with fewer bacteria [28]. Immune reconstitution with ART in the setting of untreated TB may lead to increased bacillary burden and clinically apparent disease after ART initiation. Again, in the rabbit model, the number of organisms in the rabbits with multifocal caseous necrosis and inflammation consistent with IRIS was higher than that in rabbits without evidence of IRIS. Among the 6 patients with unmasked pulmonary TB in the literature for whom microbiologic data were reported, all 5 patients who had sputum smears examined were smear and culture negative for AFB before ART and then were smear and/or culture positive after ART (present article and [2, 5, 16]). Active disease with increased bacillary burden has possible implications for increased \( M. \text{tuberculosis} \) transmission if left undiagnosed, especially in settings in which TB is less common. In resource-limited settings, patients who initiate ART often have more frequent clinical follow-up during the first few months after ART, increasing the likelihood that unmasked TB-IRIS will be promptly recognized and treated and thereby limiting the risk of ongoing transmission.

The pathogenesis of IRIS is the subject of intense study. In individuals infected with \( M. \text{tuberculosis} \), the progression of HIV-related immune dysfunction produces poor activation of CD4 T cells against specific tuberculin antigens, resulting in anergy to purified protein derivative and impaired granuloma formation. HIV-infected individuals with \( M. \text{tuberculosis} \) infection are at increased risk of progression to active disease but may manifest few specific features suggestive of active TB, thus potentially affecting accurate diagnosis especially in patients in resource-limited settings (with the exception of lymphatic TB). After the initiation of ART, memory CD4 T cells that are sequestered in lymphoid tissues are mobilized, resulting in an initial rapid increase in the number of circulating cells [41]. The combination of decreasing viral load [42] and mobilization of previously formed memory cells allows for the restoration of a specific response against mycobacterial antigens, one that can be a rapid, inflammatory, and necrotizing delayed-type hypersensitivity response with granulomatous inflammation and suppuration [26, 43]. T suppressor cells and the balance between effector and suppressor cell-mediated responses have been postulated to play an important role in the development of IRIS [44].

**Isoniazid and ART coadministration to prevent active TB and transmission.** The burgeoning problem of the occurrence of TB soon after the initiation of ART and unrecognized active disease with the risk of subsequent transmission has focused attention on empiric or preventive isoniazid therapy at the time of ART initiation. Preventive isoniazid therapy is recommended for patients coinfected with \( M. \text{tuberculosis} \), but implementation is not widespread [45–47]. In a study by Golub et al. [48], isoniazid in addition to ART decreased the risk of TB to 0.80 cases per 100 person-years, compared with 4.01 cases per 100 person-years in those receiving neither treatment, with the greatest benefit of dual therapy realized by patients with the most severe immunosuppression.

In practice—especially in resource-limited settings—the ability to conclusively rule out active TB is difficult, and the spectrum from latent to active disease is a continuum that is confounded by constitutional symptoms that could be secondary to AIDS or other atypical symptoms that are often not referable to the lungs. Extrapulmonary and/or disseminated TB may also be difficult to diagnose via sputum smear and culture. There are 2 concerns regarding the simultaneous administration of isoniazid and ART. First, if TB cannot be diagnosed by sputum smear (as is the case in >50% of HIV-infected patients), there is a risk of treating patients with active TB who may have a high enough bacillary burden to select for isoniazid resistance. Several studies have shown that the selection of isoniazid-resistant mutants is rare in patients with low bacillary burdens [49]. However, in active disease (and in some cases of subclinical TB) the rate of selection of drug-resistant mutants would be increased because of higher bacillary burdens. A mathematical model of the impact of community-wide implementation of isoniazid therapy for patients coinfected with HIV and TB bacilli showed that the incidence of TB would decrease in the short term but that rates of isoniazid-resistant TB would increase unless significant efforts to diagnose and treat active drug-sensitive and drug-resistant TB were implemented at the same time [50]. Second, diagnostic testing in patients with AIDS is challenging. Patients with the lowest CD4 T cell counts are also the most likely to be anergic to tuberculin skin testing. Sputum smear alone has poor diagnostic sensitivity in HIV-positive patients and will not identify many cases of active TB before isoniazid initiation. Patients with AIDS (i.e., those with the lowest CD4 T cell counts) are also the most likely to develop unmasked TB-IRIS after the addition of ART to antituberculous treatment [31, 51].

**Conclusion.** The present case report and review of the literature suggest that ART-associated TB occurs most often during the first 3 months after ART initiation. More-aggressive efforts to secure a diagnosis of TB before the initiation of ART as well as close monitoring after ART initiation will be important in minimizing \( M. \text{tuberculosis} \) transmission from the subset of patients in whom TB is unmasked in the setting of immune reconstitution. Prospective studies to identify patients most likely to have unrecognized TB before ART initiation could help direct empiric antituberculous chemotherapy. Only a proportion of these patients with unmasked TB will develop unmasked TB-IRIS; those with the greatest immunosuppression at the time of ART initiation, the fastest viral load suppression after ART initiation, and the highest TB antigen/organism burden are the most likely to develop IRIS. Initiation of ART at higher CD4 T cell counts will potentially decrease the incidence of unmasked TB during the first 3 months after ART, because bacillary burdens in reactivation TB will be lower. The discovery of improved diagnostics
for active TB that are both sensitive and specific remains an important priority in order to decrease the reservoir of patients with subclinical TB before ART initiation and to diagnose unmasked TB early.

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References


