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BRIEF REPORT

Cryptococcal Meningitis and Tuberculous Meningitis Co-infection in HIV-Infected Ugandan Adults

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We report 5 HIV-infected Ugandan adults with cryptococcal and tuberculous (TB) meningitis co-infection. All unmasked meningitis occurred within 5 weeks of starting HIV therapy. Xpert MTB/RIF Ultra facilitated prompt diagnosis; however, 60% in-hospital mortality occurred. TB meningitis coinfection prevalence was 0.8% (5/586) among cryptococcal meningitis, 2 during second cryptococcal episodes.

Keywords. case series; cryptococcal meningitis; HIV/AIDS; tuberculosis; tuberculous meningitis.

Meningitis remains a major cause of mortality in Africa and is the medical condition associated with highest risk of inpatient death [1]. Cryptococcal meningitis (CM) is the most common causative pathogen in advanced HIV; in 2014, there were an estimated 250 000 incident cases of cryptococcosis, accounting for 15% of AIDS-related deaths [2]. Tuberculous meningitis (TBM) is the second most common cause of HIV-associated meningitis [3]. Duel co-infection is rarely described.

In a systematic review of TB/cryptococcosis infection in China, Fang et al. identified 197 cases of co-infection, of whom 19% (n = 37) were in HIV-infected patients. TBM/CM was the most frequent co-infection (54%, 94/174) [4]. CM/TBM co-infection has been described in case reports in patients with a range of underlying immunodeficiencies, including HIV (n = 3) [5–7], systemic lupus erythematosus (n = 1), Waldenstrom's macroglobulinemia (n = 1), and reticulum cell sarcoma (n = 1), and in patients with no underlying immunodeficiency (n = 2) [8]. Here we present a case series of 5 Ugandan adults with HIV-associated CM/TBM co-infection.

METHODS

Five cases of CM/TBM co-infection were observed in the ASTRO-CM trial, a phase III, placebo-controlled, randomized clinical trial to evaluate whether sertraline, when added to standard amphotericin-based therapy for cryptococcal meningitis, leads to improved survival (ClinicalTrials.gov: NCT01802385) [9]. The primary outcome was 18-week survival. HIV-infected adults (age ≥18 years) presenting with CM to Mulago National Referral Hospital, Kampala, or Mbarara Regional Referral Hospital received standard amphotericin-based induction therapy and were randomized to receive either sertraline (400 mg/d) or placebo. CM was diagnosed by cerebrospinal fluid (CSF) using cryptococcal antigen lateral flow assay (CrAg LFA, IMMY, Norman, OK). Makerere Microbiology Laboratory performed CSF testing of white cell count, protein, and quantitative cryptococcal cultures. CSF glucose testing was mostly unavailable.

Participants were not routinely screened for CM/TBM co-infection; however, when concurrent TBM infection was suspected, participants were evaluated for TB at the physician's discretion. CSF testing for TBM was as follows: CSF Acid Fast Bacilli smear using Ziehl-Neelsen stain (Mulago Hospital only), Xpert MTB/Rif (2013–2016) and/or Xpert MTB/Rif Ultra testing (2017; Cepheid, Sunnyvale, CA), and/or for CSF Mycobacteria Growth Inhibitor Tube culture (MGIT, Becton Dickinson, Franklin Lakes, NJ). A diagnosis of CM/TBM co-infection was established in patients with microbiologically confirmed CM via a positive CSF CrAg and TBM via positive CSF Xpert MTB/Rif or MGIT culture. The cases were classified as (i) concurrent co-infection, indicating that the 2 infections were diagnosed simultaneously, or (ii) sequential co-infection when TBM was diagnosed during recurrent meningitis.

The ASTRO-CM trial protocol was approved by Ugandan and Minnesotan institutional review boards. All participants (or surrogates in cases of incapacity) provided written informed consent.

Case Presentations

From March 2015 until September 2017, 586 HIV-infected adults were diagnosed with cryptococcal meningitis among 839 consented participants with suspected meningitis. A total of 5 cases of CM/TBM co-infection were diagnosed (Table 1). All cases were confirmed TBM: 4 by Xpert MTB/RIF Ultra and 1 by culture.

The median age (range) was 32 (26–40) years, and 80% were male (4/5). All participants had advanced HIV/AIDS with a median CD4 (range) of 10 (1–79) cells/μL and a median hemoglobin of 12.2 (8.2–18.7) g/dL. All 5 were on antiretroviral

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therapy (ART) at hospital presentation. The median duration on ART at recruitment was (range) 13 (11–35) days. Two patients had no prior TB history or therapy. One patient (case 3) was on antituberculous therapy, and 2 patients (cases 1 and 4) had a previous history of treated TB infection.

Participants presented with features typical of subacute meningitis: all patients reported headache, with a median duration (range) of 30 (7–30) days; 60% (3/5) reported fever, 40% (2/5) reported seizures, and 20% (1/5) reported visual disturbance or photophobia, respectively. The median Glasgow Coma Score (GCS) was (range) 12 (6–15), and 40% (2/5) of patients had a sixth cranial nerve palsy on examination. CSF opening pressure at diagnosis was raised (>20 cmH_20) in 40% (2/5), with a median opening pressure of (range) 16 (5–46) cmH_20. The baseline median CSF white cell count (range) was 60 (<5–2200) cells/μL, and median protein was 100 (20–356) mg/dL. Two patients (40%) had sterile CSF cryptococcal cultures with positive serum and CSF CrAg tests.

TBM was diagnosed concurrently in 3 cases and sequentially in 2 cases (cases 1 and 2). Case 1 had a previous history of CM, completing treatment 12 years earlier, and presented on this occasion 1 month after restarting ART with a 1-month history of meningitis symptoms. At recruitment, the CD4 was 10 cells/μL, CSF CrAg LFA was faintly positive, CSF white cells <5 cells/μL, and CSF protein 20 mg/dL. Antifungal treatment was started for possible cryptococcal relapse, although CSF fungal cultures remained sterile. A second lumbar puncture on day 3 of antifungal therapy revealed a CSF white cell count of 165 cells/μL (100% lymphocytes), with protein of 74 mg/dL. The participant died on day +10 before mycobacterial cultures grew on day +11. Case 2 presented to the hospital 173 days after his initial cryptococcal diagnosis with recurrence of meningitis symptoms and was diagnosed with TBM promptly by Xpert Ultra.

The in-hospital mortality was 60% (3/5). One patient was alive at 18-week follow-up. Three patients died 2–11 days after diagnosis. The fifth patient survived to hospital discharge, but they were not actively followed thereafter. Four CM/TBM diagnoses were made after January 2017, when Xpert MTB/RIF Ultra testing was introduced. Before this time, 0.2% (1/453) with confirmed CM were diagnosed with concurrent TBM, compared with 3% (4/133) in the post–Xpert MTB/RIF Ultra period ($P = .011$).

DISCUSSION

While CM/TB co-infection has been previously described [10, 11], CM/TBM co-infection is reported to be extremely rare. The diagnosis of CM/TBM co-infection is particularly challenging due to the paucibacillary nature of TBM. As such, data on HIV-associated CM/TBM co-infection are limited to individual case reports [5–7]. The introduction of Xpert MTB/Rif, which is a cartridge-based, fully automated polymerase chain reaction molecular assay, was a step forward. However, the introduction in 2016 of the re-engineered Xpert MTB/RIF Ultra has significantly improved our TBM diagnostic capacity. In the context of these new diagnostics, we report a case series of 5 HIV-associated CM/TBM co-infection cases.

In our cohort of severely immunocompromised HIV-infected adults with cryptococcosis, where TBM was selectively screened for, CM/TBM coinfection occurred in 0.5% (3/586) of first episodes of cryptococcosis and 0.8% (5/586) overall, with 2 sequential TBM infections being recurrent meningitis episodes. Our Ugandan prevalence is lower than the 1.5% CM/TBM co-infection rate reported among HIV-associated CM patients (8/514) in South Africa, where the TB incidence is ~5-fold higher [12]. A single-center case series from Taiwan [11] reported 23 cases of cryptococcosis/TB co-infection at any

Table 1. Clinical Characteristics of 5 Cases of HIV-Associated Cryptococcal Meningitis and Tuberculous Meningitis Co-infection

Case	Age	Sex	Days on ART	CD4 Cells/μL	History of CM	History of TB	Days From CM to TBM Diagnosis	CSF TB Diagnostics	CSF White Cells/μL	CSF Protein, mg/dL	*C*ryptococcus CFU/mL CSF	Outcome
1	40	F	30 10	12 y prior	Y	11	Xpert MTB/RIF (-) MGIT culture (+)	<5 155[a]	20 74[a]	0 0[a]	Death, day 10
2	35	M	13 186[b]	79	N	N	173	Xpert Ultra (+) MGIT culture not done	115 2200[b]	157 356[b]	12 300 0[b]	Alive at discharge
3	26	M	35	19	N	N	0	Xpert Ultra (+) MGIT culture (-)	205	100	0	Death, day 11
4	32	M	11	1	N	Y	0	Xpert Ultra (+) MGIT culture contaminated	210	40	520 000	Death, day 2
5	32	M	5	3	N	N	0	Xpert Ultra (+) MGIT culture not done	<5	N/A	20 000	Alive >18 wk

Abbreviations: ART, antiretroviral therapy; CFU, colony-forming units; CM, cryptococcal meningitis; CSF, cerebrospinal fluid; MGIT, mycobacteria growth indicator tube; TB, tuberculosis; TBM, tuberculous meningitis.

[a]Day 3 lumbar puncture.

[b]At the time of TBM diagnosis.

site (11/23, 48% in HIV-infected patients), representing 5.4% of cryptococcosis cases.

The marked overlap in clinical presentation between CM and TBM may lead to delayed or missed diagnoses. Indeed, the clinical presentations and CSF profiles observed in our CM/TBM co-infection cohort are comparable to those described in HIV-associated CM or TBM mono-infection. The high early mortality rate associated with CM may also mean that CM patients die before a diagnosis of concurrent TBM is made, which may also result in underdiagnosis of co-infection. To date, there are no discriminatory biomarkers that can distinguish co-infection from CM or TBM mono-infection.

CM/TBM co-infection reflects the advanced immunosuppression characteristic of patients with HIV-associated CM and the complexities of diagnosis and management in patients who remain at risk of intercurrent opportunistic infections. This case series demonstrates both the need to consider co-infection at baseline diagnosis and the need to remain vigilant for co-infection throughout the follow-up period.

Each case of CM/TBM co-infection occurred in patients who had recently started on ART (median time on ART, 13 days). This contrasts with the 44% (202/460) of the overall ASTRO cohort who were receiving ART at recruitment. Although our numbers are small, this may suggest that patients presenting with CNS infection soon after ART initiation, a clinical phenotype consistent with unmasking immune reconstitution inflammatory syndrome (IRIS), may be at particularly high risk of CM/TBM co-infection. As demonstrated by cases 1–2, considering TBM is important in patients presenting with suspected CM relapse following ART initiation. IRIS is caused by recovery of pathogen-specific immune responses following ART initiation that result in a pathological inflammatory response. In unmasking CM-IRIS, this manifests as a patient with previously undiagnosed cryptococcosis presenting with meningitis soon after ART is started. In unmasking TBM-IRIS, dormant Rich foci on the meninges become inflamed due to TB-specific immune responses, resulting in rupture and translocation of bacilli into the subarachnoid space, with associated inflammatory meningitis. Pathogen-specific immune responses may recover simultaneously or sequentially, resulting in IRIS phenomena to several pathogens and therein the presentation of co-infections.

In conclusion, CM/TBM co-infection in HIV-infected adults remains a rare condition, but one of which physicians should remain aware. The introduction of molecular tests like Xpert MTB/Rif Ultra has improved our capacity to diagnose TBM; whether patients presenting with CM unmasking IRIS are at higher risk of CM/TBM co-infection and whether co-infection is associated with increased mortality warrant further investigation. Providers should be aware that the recent rapid upscaling of "HIV test and treat" could result in an increased incidence of co-infection unmasking IRIS events.

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