Published in final edited form as:

Curr Opin Infect Dis. 2016 February ; 29(1): 10–22. doi:10.1097/QCO.0000000000000224.

The Immunopathogenesis of Cryptococcal Immune Reconstitution Inflammatory Syndrome - Understanding a Conundrum

David B Meya^{1,2,3}, Yukari C Manabe⁴, David R Boulware², and Edward N Janoff⁵

¹Infectious Disease Institute, Makerere University, Uganda ²Dept of Medicine, Center for Infectious Diseases and Microbiology Translational Research, University of Minnesota, USA ³School of Medicine, College of Health Sciences, Makerere University ⁴Division of Infectious Diseases, Department of Medicine, Johns Hopkins University, Baltimore, MD ⁵Mucosal and Vaccine Research Program Colorado (MAVRC), University of Colorado Denver, Aurora, Colorado, USA; Denver Veterans Affairs Medical Center, Denver, CO

Abstract

Purpose of Review—Cryptococcal meningitis (CM) causes significant mortality among HIV-infected patients, despite antifungal therapy and use of antiretroviral therapy (ART). In patients with CM, ART is often complicated by immune reconstitution inflammatory syndrome (IRIS), manifesting as unmasking of previously unrecognized subclinical infection (unmasking CM-IRIS) or paradoxical worsening of symptoms in the central nervous system after prior improvement with antifungal therapy (paradoxical CM-IRIS). We review our current understanding of the pathogenesis of this phenomenon, focusing on unifying innate and adaptive immune mechanisms leading to the development of this often fatal syndrome.

Recent Findings—We propose that HIV-associated CD4⁺ T cell depletion, chemokine-driven trafficking of monocytes into cerebrospinal fluid (CSF) in response to CM, and poor localized innate cytokine responses lead to inadequate cryptococcal killing and clearance of the fungus. Subsequent ART-associated recovery of T cell signaling and restored cytokine responses, characterized by Interferon- γ production, triggers an inflammatory response. The inflammatory response triggered by ART is dysregulated due to impaired homeostatic and regulatory mechanisms, culminating in the development of CM-IRIS.

Summary—Despite our incomplete understanding of the immunopathogenesis of CM-IRIS, emerging data exploring innate and adaptive immune responses could be exploited to predict, prevent and manage CM-IRIS and associated morbid consequences.

Keywords

Cryptococcus; HIV; IRIS; Immunopathogenesis; Cryptococcal Meningitis

Corresponding Author: David B Meya MBChB, MMed, Infectious Disease Institute, #22418, College of Health Sciences, Makerere University, Kampala, Uganda. Phone: +256 312 307 224; david.meya@gmail.com.

Conflict of Interest

All authors have no conflict of interest to declare.

Introduction

Cryptococcal meningitis (CM) causes 20–25% of AIDS-related mortality worldwide [1]. In sub-Saharan Africa, *Cryptococcus* is the most common cause of meningitis in adults, accounting for 26% of cases in Malawi, 45% in Zimbabwe, 30% in South Africa, and 60% in Uganda [2–6]. Despite antiretroviral therapy (ART) scale up, many patients with a new HIV diagnosis still present with very low CD4⁺ T cells (<200/μL) [7] and opportunistic infections, such as Cryptococcal meningitis, due to failure to initiate ART in a timely manner and poor linkage to HIV care.

Initiation of ART can be complicated by Immune reconstitution inflammatory syndrome (IRIS), which contributes to the mortality following ART. IRIS develops in up to 30% of patients with CM, manifesting as clinical deterioration a few weeks or months after initiating ART [8, 9]. This syndrome is thought to be an aberrant inflammatory response to persistent cryptococcal antigens during recovery of the host immune system [10]. IRIS may manifest as unmasking of previously unrecognized subclinical infection (unmasking CM-IRIS) or paradoxical worsening of CM-associated symptoms in the central nervous system after improvement with antifungal therapy (paradoxical CM-IRIS). Occurring within the closed central nervous system space, CM-IRIS can be fatal, and has been associated with a high mortality (Table 1).

Role of Primary Immune or Host response

Primary immune deficiencies of CD4⁺ T cells, impaired phagocytic function of macrophages, and variants in major histocompatibility complex molecules I/II are associated with development of serious cryptococcal infections [23, 24]. Similarly, patients with X-linked hyper-IgM and hyper-IgE syndromes, and those with secondary immune disorders, such as malignancies, solid organ transplants, use of immunosuppressant drugs, and with HIV infection show a similar predisposition [25, 26]. In healthy humans, the initial immune response involves fungal recognition by lung innate immune cells and generation of signals that lead to expansion of *Cryptococcus*-specific CD4⁺ T cells to control the infection [27]. Minimal trafficking of immune cells to the CSF occurs in severely immunosuppressed HIV-infected patients with CM [28*]. The primary immune response to cryptococcal infection may determine the mechanisms that eventually cause cryptococcal IRIS.

In the brain, Glucuronoxylomannan, (GXM), the principal component of the cryptococcal polysaccharide capsule, binds to macrophage FcγRIIIB, which inhibits the Fcγ receptor and proinflammatory cytokine expression [29, 30], whereas mannoprotein induces antigen-specific T cell responses [31]. *C. neoformans* promotes an imbalance toward a Th2 response by suppressing T-helper (Th) 1 responses [32–34]. Uncontrolled murine cerebral cryptococcosis results from IL-4/IL-13-dependent alternative activation of macrophages [35]. Consistent with these observations, patients who later develop CM-IRIS show higher baseline serum levels of Th2 cytokines (IL-4 and IL-13), lower levels of Tumor Necrosis Factor (TNF)-α [19] and lower CSF levels of IFN-γ [36] at CM diagnosis compared with those who do not develop CM-IRIS.

Cryptococcus inhibits T cell migration into the CSF by increasing L-selectin shedding from T cells, limiting their ability to extravasate into the CNS [37, 38]. The imbalance of these functional immune constituents may underlie the pathologic clinical outcome of ART-induced IRIS during cryptococcal infection. In patients with AIDS, *Cryptococcus* establishes a chronic infection. Upon ART-associated immune recovery, the persistent fungal antigen burden is proposed to trigger an exaggerated inflammatory response due to HIV-associated impairment of homeostatic regulatory mechanisms [39].

Immune Reconstitution with Antiretroviral Therapy

During early HIV infection, HIV-specific immune responses limit viral replication and the destruction of CD4⁺ T cells is balanced by the generation of new CD4⁺ T cells [40]. Over time, ongoing viral replication, immune cell depletion and dysregulation are associated with chronic inflammation, resulting in scarring and destruction of the normal lymph node architecture [41]. ART suppresses HIV replication and CD4⁺ T cell apoptosis, allowing immune regeneration. During immune reconstitution, CD4⁺ T cells increase rapidly (26 cells/mm³/month) [42], some pathogen-specific immune responses recover [43*], a decreased susceptibility to OIs is observed, and genes encoding innate antiviral responses, immune activation, cellular proliferation and apoptosis are downregulated [44]. HIV infection also affects the innate immune axis, with expansion of monocytes [45], whereas immune reconstitution decreases the activation of monocytes/macrophages [46].

Epidemiology of Cryptococcal Meningitis Immune Reconstitution Inflammatory Syndrome (CM-IRIS)

An estimated 10%–25% of persons without a known OI who start ART experience IRIS [40, 47–49]. IRIS has been associated with >20 infectious pathogens (*Mycobacterium avium* complex, *M. tuberculosis*, *Cryptococcus neoformans*, Cytomegalovirus, JC virus, *Pneumocystis jirovecii*, Herpes zoster (VZV), and hepatitis B and C), malignancy, (Kaposi's sarcoma) and occasionally non-infectious autoimmune syndromes and malignancies [40, 41, 47, 48]. The majority of IRIS events will occur within the first eight weeks of ART, however, delayed IRIS occurring years later has also been described, particularly for cryptococcal-IRIS and CMV-IRIS [11, 50].

Approximately 25% of CM patients treated with antifungals and ART [8, 9, 16] will experience paradoxical deterioration, despite decreasing fungal burden [8, 16, 19, 51] (Table 1). Cryptococcal IRIS may manifest as relapsing aseptic meningitis, increased intracranial pressure, new focal neurologic signs, intracranial cryptococcomas, lymphadenopathy, and/or development of abscesses [11, 19, 52–54] which can present as multifocal or multiphasic disease [55*]. Mortality associated with cryptococcal IRIS ranges from 0 to 67% [19, 20, 50].

In the absence of HIV coinfection, cryptococcal IRIS has also been described with reversal of iatrogenic immunosuppression. Cutaneous cryptococcal IRIS was reported in a patient with T cell pro-lymphocytic leukemia [56] following use of alemtuzumab, a monoclonal antibody that blocks CD52, a surface ligand involved in immune cell mobility [57]. Tumor

necrosis factor (TNF)- α inhibitors have also been associated with an increased risk of cryptococcal infections [58–60], highlighting the important role played by TNF- α in the immune response against *Cryptococcus* [61].

Case Definition- Cryptococcal IRIS

The International Network for the Study of HIV-associated IRIS (INSHI) has developed a consensus case definition for CM-IRIS [51] (Supplementary Table 1). IRIS remains a diagnosis of exclusion, after considering adverse drug reactions, new OIs, and microbiologic treatment failure (whether due to drug-resistance or malabsorption). Differentiation of IRIS from these other diagnoses has important implications that may impact clinical management and outcomes.

Clinical Risk factors for Cryptococcal IRIS

The immune response to *Cryptococcus* requires both the innate and adaptive immune axes to be functionally coupled to control the infection [62*, 63]. During HIV infection, the adaptive immune response is severely compromised and fungal dissemination to the meninges occurs in the absence of antifungal therapy. ART alone is insufficient to prevent cryptococcal disease when subclinical infection is present [64]. Microbiologic, virologic, immunologic and therapeutic factors may affect the incidence of CM-IRIS (Table 2).

Proposed immunologic mechanisms for CM-IRIS

We highlight three stages of CM-IRIS pathogenesis (Table 3): 1) pre-ART paucity of inflammation, immune system dysfunction, and/or anergy resulting in poor macrophage killing and clearance of cryptococcal organisms; 2) macrophages are poised to receive missing T cell signals, such as from IFN- γ , which are restored after ART initiation and induce cryptococcal killing triggering an inflammatory response; and 3) dysregulated homeostatic and regulatory mechanisms leading to exaggerated responses.

Phase 1: Paucity of inflammatory response

Neutrophil migration and fungistatic activity is impaired during HIV infection [74, [Walenkamp, 2003 #299]]. Poor CSF cytokine responses predispose to poor antigen clearance and increase the risk of IRIS. At CM diagnosis, 3 of 5 studies suggest a paucity of inflammation increases the risk of developing CM-IRIS (Table 4a). Significant elevation of cells and protein is not consistently demonstrated. HIV-uninfected subjects with CM show a more robust immune response in CSF. Patients with HIV infection had higher cryptococcal antigen titers at baseline, with a significant decrease at CM-IRIS. It remains unclear whether the changes that occur at CM-IRIS suggest a recovering immune system responding normally or reflect exaggerated immune responses to residual cryptococcal antigen. The recurrence of symptoms/signs of meningitis associated with CM-IRIS in the setting of negative CSF cultures, however, appears to fit the latter.

Table 4b summarizes studies that have measured cytokines/chemokines in blood and CSF among patients with CM or CM-IRIS. These data suggest that CM patients with decreased

innate (IL-6, IL-8, TNF- α) and adaptive inflammatory cytokines (IFN- γ), at CM diagnosis are at higher risk of developing CM-IRIS. Elevation of the inflammatory cytokines, Th1 (IFN- γ), Th17 (IL-17) and the innate cytokines (TNF- α) occurs during CM IRIS. Notably, two studies suggest that increased expression of the chemokines, CCL2 and CCL3 at CM diagnosis is associated with risk of CM-IRIS [71, 80*]. Evaluating immunologic changes concurrently in CSF and blood sampled at CM diagnosis, after ART initiation, at CM-IRIS and after the IRIS event from patients with CM-IRIS compared with matched controls without CM-IRIS would identify the immunologic signature that defines the risk and immunopathogenesis of CM-IRIS.

Naranbhai et al show during CM, natural killer (NK) cells are involved in CNS immune regulation during the primary response to cryptococcal infection via elevated expression of the chemokines CXCR3, CX3CR1 and diminished TNF- α in CSF [79*]. We have demonstrated elevated frequencies of TNF- α - and IL-6-producing monocytes at CM-IRIS compared with ART matched controls without CM-IRIS [73].

The timing of ART initiation in relation to diagnosis and therapy of CM may affect the incidence of CM-IRIS [8, 18, 19, 65, 67, 68*, 70], however, the increased incidence of cryptococcal IRIS with earlier ART initiation has not been consistent across cohorts. The excess deaths occurring with earlier ART initiation may be distinct IRIS events although clinically indistinguishable from deterioration due to cryptococcosis.

Phase 2: Innate cell activation

Wiesner and Boulware propose that in the early stages of ART immune recovery, in the absence of an effective T cell response, foreign antigens generate ongoing pro-inflammatory signaling by antigen presenting cells, demonstrated by increasing serum IL-6 and C reactive protein in the weeks prior to the IRIS event [19, 81]. The corresponding effector response is delayed until a cytokine storm predominated by a Th1 response occurs at the time of IRIS in blood and CSF [19, 36]. Similarly, Barber and colleagues suggest that, prior to ART, killing of intracellular pathogens by innate phagocytes is defective in the absence of IFN- γ signaling from CD4⁺ T cells [82**]. They further propose that partially primed myeloid cells accumulate with disease progression and the creation of a hyperresponsive state of the innate immune cells. Upon immune reconstitution of CD4⁺ T cell help with ART, broad activation of these myeloid cells results in the production of pro-inflammatory mediators, eliciting the immunopathology defining IRIS. Thus, IRIS is thought to result from uncoupling of the innate and adaptive immune axes following repletion of HIV-associated CD4⁺ T cell number and function with ART.

Differences in chemokine and cytokine expression occur in CSF and blood compartments pre-ART and during CM-IRIS with increased expression of CCL2 (monocyte chemo attractant protein-1, MCP-1) and CCL3 (macrophage inflammatory protein 1- α , or MIP 1- α) [80*]. These chemokines recruit myeloid cells into the CNS, suggesting monocyte recruitment into the CSF occurs during CM-IRIS. CCL2 and CCL3 were also higher in CSF of patients who remained culture positive following antifungal therapy, reflecting ongoing innate antigen presenting cell signaling among this culture-positive group with a higher risk

of developing CM-IRIS. More recently, Jarvis et al have demonstrated that elevated CCL2 and CCL3 levels in CSF, low peripheral CD4⁺ T cells and low CSF white cell numbers at CM diagnosis were predictive of CM-IRIS [71]*. The authors suggest that over recruitment of myeloid cells into the CNS could lead to exaggerated immune responses that drive the immunopathology observed during CM-IRIS. Three subsets of monocytes are known to be present in blood depending on their degree of expression of the CD14 surface marker: classical monocytes (CD14⁺⁺ CD16⁻), intermediate monocytes (CD14⁺⁺CD16⁺) and the non-classical monocyte (CD14⁺CD16⁺⁺ monocyte) [83]. We have recently demonstrated differences in phenotype and activation of monocytes and CD4⁺ T cells in blood compared to CSF during CM-IRIS, with the more pro-inflammatory intermediate monocytes predominating in the CSF during CM-IRIS [28*]. Additionally, mortality in the early initiation of ART arm of the Cryptococcal Optimal ART Timing (COAT) trial was associated with elevated CCL2/CCL3 and macrophage/microglial activation in the CSF [84*]. We surmise that localized immune responses, particularly monocyte innate immune responses, are involved in the immunopathology during CM-IRIS.

Phase 3: Immune dysregulation

Almost every constituent of the Immune system is deranged during very advanced HIV infection, the time at which CM occurs (CD4⁺ T cells 15–30/uL).

Regulatory T cells

Regulatory T cells (Treg) downregulate immune responses to foreign antigens to prevent collateral damage from inflammatory responses [39]. However, a dampened immune response could also have the detrimental effect of delaying antigen clearance during acute infection [85]. A decrease in regulatory T cells at IRIS is thought to predispose to dysregulated effector responses. A higher proportion of regulatory T cells among subjects who developed CM-IRIS compared to healthy controls at baseline was found in one study [86] while substantial quantitative expansion of these cells with defective immunosuppressive function was found in another study during IRIS [87]. Several studies show no association between decreased proportions of regulatory T cells, including the T effector/T regulatory ratio, and IRIS [86, 88, 89] suggesting that these cells may have a limited role in the pathogenesis of CM-IRIS, however, anti-TNF- α molecules in cryptococcal IRIS enhance Treg and Th2 predominance over Th1 responses [90*]. Further studies focusing on the functional role of regulatory T cells during CM-IRIS are warranted.

Innate Immune Axis

Gene expression signatures related to infection and inflammation in circulating monocytes prior to ART initiation and at TB IRIS [91*, 92] demonstrate that increased numbers and activation of the classical monocyte subset appear to functionally predispose to and mediate the development of TB-IRIS with increased expression of IL-1 β , IL-6 and TNF during TB-IRIS [93**]. Monocytes represent a smaller minority population in the CSF (8–10% of mononuclear cells [46, 94]) in healthy adults. However, during CM-IRIS, CSF monocyte subsets shift from classical to an intermediate/pro-inflammatory subset, with elevated programmed death ligand-1 (PD-L1) expression on monocytes and NK cells [28*]. The

absence of non-classical monocytes in patients with CM pre-ART is associated with the development of CM-IRIS possibly due to poor fungal clearance. Notably, robust IL-6 and TNF- α responses to IFN- γ stimulation *ex vivo* occurred in patients experiencing CM-IRIS compared to controls [73*]. Two case reports showed improvement of cryptococcal IRIS in a solid organ transplant recipient and in an HIV-infected patient with an IRIS-related cryptococcoma after using adalimumab, which blocks human TNF- α [95*, 96]. Taken together, these data corroborate involvement of the innate immune axis in cryptococcal IRIS.

Lymphopenia-induced proliferation

Space-driven homeostatic compensatory T cell proliferation occurring soon after ART initiation could link the innate and adaptive immune responses that are observed at CM-IRIS. Lymphopenia, such as occurs in HIV infection drives the homeostatic proliferation of memory-like T cells [97, 98]. Spontaneous proliferation could be induced by high-affinity major histocompatibility complex (MHC)/peptide interactions occurring in the presence of residual cryptococcal antigens and monocyte-derived IL-6 [99–102]. Elevated IL-6 and TNF- α -expressing monocytes pre-ART [73*] and an increase in IL-6 prior to development of CM-IRIS [19] appear to confirm a role for IL-6. Spontaneous proliferation causes naïve T cells to acquire the phenotypic and functional properties of effector memory cells [103] that upregulate IFN- γ expression and related Th1 responses that predominate during CM-IRIS as the state of immune suppression is reversed. Additionally, IL-2, IL-4, IL-7 and IL-15 favor memory T cell survival and expansion [87, 104, 105]. It is plausible that IL-6 produced by monocytes drives the proliferation and expansion of antigen specific CD4⁺ T cell memory cells which then provide signaling that fully activates macrophages primed by *Cryptococcus* in the brain parenchyma, with a resultant spike in inflammation in CSF during CM-IRIS.

That T cells play a pathogenic role in CM-IRIS is supported by both indirect evidence from cytokine studies both in blood and CSF, prior to ART initiation and at the time of IRIS [19, 36] as well as direct evidence of an influx of CD4⁺ T cells into the CSF at the time of IRIS [28]. The restoration of memory T cell responses coupled with dysregulated memory responses against existing residual antigen have been considered the source of this exuberant secretion of cytokines resulting in a ‘cytokine storm’ proposed to underlie CM-IRIS [106]. However, the degree of inflammation present in the CSF at time of CM-IRIS for such a ‘cytokine storm’ is not supported by the literature (Tables 4a and 4b). Rather, cellular and cytokine responses in CSF are actually quite similar to the cytokines levels present during initial CM among non- CM-IRIS patients [16, 36]. One potential confounding issue is that just as results in blood likely do not accurately reflect processes in the CSF, CSF may not be the best monitor of IRIS pathology but is merely the most accessible. IRIS pathology may be occurring primarily in the brain tissue, for which CSF is only a surrogate. Autopsy studies may help to clarify these compartmental differences, including the idea that during CM-IRIS, highly differentiated polyfunctional antigen-specific CD4⁺ T cells are expanded, and less so, the CD8⁺ T cells [106].

Effector T cells and Cytokine responses

Dysregulated expansion of activated antigen-specific T cells as severe immune suppression reverses following ART initiation is thought to drive CM-IRIS. A functionally impaired yet predominantly highly activated CD4⁺ T cell effector memory phenotype in blood is characteristic of patients with IRIS prior to and during IRIS episodes [89]. Pre-ART, programmed death-1 (PD-1), an inhibitory receptor that is up-regulated on T cells following antigen stimulation, was elevated on both CD4⁺ and CD8⁺ T cells among subjects who developed IRIS compared with non-IRIS controls [89], suggesting defects in T cell inhibitory control [89]. Further, a robust increase of polyfunctional, highly differentiated effector memory CD4⁺ T cell responses specifically against the underlying pathogen is a distinctive feature of IRIS [106], although T cells reactive with some pathogens may not recover with ART [43*].

Cytokines and chemokines measured in the CSF or blood provide insight into the mechanisms of CM-IRIS from patterns pre-ART or during IRIS events. IFN- γ largely defines Th1 responses with possible contributions from IL-2 and TNF- α whereas IL-4, IL-5, and IL-13 denote Th2 responses. A paucity of Th1 responses in CSF at CM diagnosis, thought to be associated with poor cryptococcal clearance, is considered a risk factor for developing future CM-IRIS [36]. However, an increase in the Th1 response in CSF coincides with an increase in CD4⁺ T cells in CSF with ART during CM-IRIS [28*, 36, 107]. These cytokine responses could be identified both in CSF and in peripheral blood [19, 108]. Of note, localized responses in CSF at the site of the effective response may not parallel those in blood [19, 36]. Understanding these cytokine responses could be useful in developing measurable biomarkers to stratify the risk of developing CM-IRIS and immunologic endpoints for therapeutic intervention studies to treat or prevent CM-IRIS. A proposed integrated model characterizing these features of CM IRIS pathogenesis is highlighted in Figure 1.

Future Perspectives

Despite the benefits of ART in HIV-infected patients with CM, a dysregulated immune system after ART initiation poses clinical challenges. As advances are made in understanding the mechanistic pathways for paradoxical CM-IRIS, evidence of innate immune responses, specifically monocytes in the events leading up to paradoxical CM-IRIS is emerging. Our understanding of the effector immune responses involved in the pathogenesis of unmasking CM-IRIS remains to be elucidated.

Earlier initiation of ART soon after the diagnosis of CM when antigen levels are high is associated with worse outcomes [68*]. However, these excess early deaths were not directly related to the development of paradoxical IRIS, at least using current case definitions [51]. This clinical definition may be modified to include innate and adaptive biomarkers as we develop a more mechanistic understanding of the immunopathogenesis of CM-IRIS.

Because the dysregulated expansion of antigen-specific T cells [106] during paradoxical CM-IRIS occurs in the setting of residual cryptococcal antigen, decreasing the fungal burden more rapidly with more effective fungicidal agents in the induction phase of CM treatment

is germane. Such treatments may include customized therapy using higher doses of fluconazole and a longer duration of Amphotericin or flucytosine for patients with high fungal burden, use of adjunctive interferon- γ [21], and other antifungal drugs such as sertraline [109*] to more rapidly reduce fungal burden. A more effective approach is to identify subclinical cryptococcal infection prior to ART and intervene preemptively with fluconazole, preventing CM and CM-IRIS, as recommended by the WHO [110]. Immunologic interventions to prevent and manage paradoxical CM-IRIS could inhibit monocyte migration into the CNS using CCR5 inhibitors, IL-6 blockade with siltuximab, inhibit TNF- α inhibitor with adalimumab and steroids.

Conclusions

Cryptococcal IRIS remains a conundrum, for effective prevention, diagnosis and management. A confluence of factors contribute to this syndrome, including initial HIV-associated CD4⁺ T cell depletion with attenuated localized inflammatory responses, followed by chemokine-driven trafficking of monocytes into the cerebrospinal fluid, the recovery and restoration of T cell signaling after initiating ART in the setting of dysregulated innate, adaptive and regulatory immune responses. Our current understanding of CM-IRIS provides further insight into how these cellular interactions could be exploited to predict, prevent and manage CM-IRIS.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We would like to acknowledge Prof. Paul Bohjanen for his insightful and critical review of the manuscript.

Financial support and sponsorship

This work supported by the National Institutes of Health (R01AI078934, U01AI089244, R21NS065713, R01AI108479, T32AI055433) to DM, DB; K24AI096925 to PB; the Wellcome Trust (Training Health Researchers into Vocational Excellence (THRiVE) in East Africa, grant number 087540 to DM; AI108479 to EJ, and the Veterans Affairs Research Service.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

*of special interest

**of outstanding interest

1. Park BJ, Wannemuehler KA, Marston BJ, et al. Estimation of the current global burden of cryptococcal meningitis among persons living with HIV/AIDS. *AIDS*. 2009; 23:525–30. [PubMed: 19182676]
2. Cohen DB, Zijlstra EE, Mukaka M, et al. Diagnosis of cryptococcal and tuberculous meningitis in a resource-limited African setting. *TM & IH*. 2010; 15:910–7. [PubMed: 20573075]

3. Hakim JG, Gangaidzo IT, Heyderman RS, et al. Impact of HIV infection on meningitis in Harare, Zimbabwe: a prospective study of 406 predominantly adult patients. *AIDS*. 2000; 14:1401–7. [PubMed: 10930155]
4. Jarvis JN, Meintjes G, Williams A, et al. Adult meningitis in a setting of high HIV and TB prevalence: findings from 4961 suspected cases. *BMC Inf Dis*. 2010; 10:67.
5. Durski KN, Kuntz KM, Yasukawa K, et al. Cost-effective diagnostic checklists for meningitis in resource-limited settings. *J Acquir Immune Defic Syndr*. 2013; 63:e101–8. [PubMed: 23466647]
6. Gordon SB, Walsh AL, Chaponda M, et al. Bacterial meningitis in Malawian adults: pneumococcal disease is common, severe, and seasonal. *Clin Inf Dis*. 2000; 31:53–7.
7. Avila D, et al. IeDea Collaborations ARTC. Immunodeficiency at the start of combination antiretroviral therapy in low-, middle-, and high-income countries. *J Acquir Immune Defic Syndr*. 2014; 65:e8–16. [PubMed: 24419071]
8. Lortholary O, Fontanet A, Memain N, et al. Incidence and risk factors of immune reconstitution inflammatory syndrome complicating HIV-associated cryptococcosis in France. *AIDS*. 2005; 19:1043–9. [PubMed: 15958835]
9. Muller M, Wandel S, Colebunders R, et al. Immune reconstitution inflammatory syndrome in patients starting antiretroviral therapy for HIV infection: a systematic review and meta-analysis. *The Lancet Inf Dis*. 2010; 10:251–61.
10. French MA. HIV/AIDS: immune reconstitution inflammatory syndrome: a reappraisal. *Clin Inf Dis*. 2009; 48:101–7.
11. Jenny-Avital ER, Abadi M. Immune reconstitution cryptococcosis after initiation of successful highly active antiretroviral therapy. *Clin Inf Dis*. 2002; 35:e128–33.
12. Lawn SD, Bekker LG, Myer L, et al. Cryptococcal immune reconstitution disease: a major cause of early mortality in a South African antiretroviral programme. *AIDS*. 2005; 19:2050–2. [PubMed: 16260920]
13. Shelburne SA 3rd, Darcourt J, White AC Jr, et al. The role of immune reconstitution inflammatory syndrome in AIDS-related *Cryptococcus neoformans* disease in the era of highly active antiretroviral therapy. *Clin Inf Dis*. 2005; 40:1049–52.
14. Jenkin, LKA. Cryptococcal complications in the first year on HAART: experiences of a South African antiretroviral programme. XVI International AIDS Conference; Toronto, ON, Canada. 2006. p. Abstract THPE0080
15. Sungkanuparph S, Jongwutiwes U, Kiertiburanakul S. Timing of cryptococcal immune reconstitution inflammatory syndrome after antiretroviral therapy in patients with AIDS and cryptococcal meningitis. *J Acquir Immune Defic Syndr*. 2007; 45:595–6. [PubMed: 17704683]
16. Bicanic T, Meintjes G, Rebe K, et al. Immune reconstitution inflammatory syndrome in HIV-associated cryptococcal meningitis: a prospective study. *J Acquir Immune Defic Syndr*. 2009; 51:130–4. [PubMed: 19365271]
17. Yan S, Chen L, Wu W, et al. Paradoxical immune reconstitution inflammatory syndrome associated with cryptococcal meningitis in China: a 5-year retrospective cohort study. *Clin Micro Inf*. 2014
18. Kambugu A, Meya DB, Rhein J, et al. Outcomes of cryptococcal meningitis in Uganda before and after the availability of highly active antiretroviral therapy. *Clin Inf Dis*. 2008; 46:1694–701.
19. Boulware DR, Meya DB, Bergemann TL, et al. Clinical features and serum biomarkers in HIV immune reconstitution inflammatory syndrome after cryptococcal meningitis: a prospective cohort study. *PLoS medicine*. 2010; 7:e1000384. [PubMed: 21253011]
20. da Cunha Colombo ER, Mora DJ, Silva-Vergara ML. Immune reconstitution inflammatory syndrome (IRIS) associated with *Cryptococcus neoformans* infection in AIDS patients. *Mycoses*. 2011; 54:e178–82. [PubMed: 20337940]
21. Jarvis JN, Meintjes G, Rebe K, et al. Adjunctive interferon-gamma immunotherapy for the treatment of HIV-associated cryptococcal meningitis: a randomized controlled trial. *AIDS*. 2012; 26:1105–13. [PubMed: 22421244]
22. Chang CC, Dorasamy AA, Gosnell BI, et al. Clinical and mycological predictors of cryptococcosis-associated immune reconstitution inflammatory syndrome. *AIDS*. 2013; 27:2089–99. [PubMed: 23525034]

23. Lanternier F, Cypowyj S, Picard C, et al. Primary immunodeficiencies underlying fungal infections. *Curr Opin Ped*. 2013; 25:736–47.
24. Antachopoulos C, Walsh TJ, Roilides E. Fungal infections in primary immunodeficiencies. *Eur J Ped*. 2007; 166:1099–117.
25. Pagano L, Fianchi L, Caramatti C, et al. Cryptococcosis in patients with hematologic malignancies. A report from GIMEMA-infection. *Haematologica*. 2004; 89:852–6. [PubMed: 15257938]
26. Baddley JW, Forrest GN. Practice ASTIDCo. Cryptococcosis in solid organ transplantation. *Am J Transp*. 2013; 13(Suppl 4):242–9.
27. Huffnagle GB, Lipscomb MF. Cells and cytokines in pulmonary cryptococcosis. *Research in Immunology*. 1998; 149:387–96. discussion 512–4. [PubMed: 9720956]
28. Meya DB, Okurut S, Zziwa G, et al. Cellular Immune Activation in Cerebrospinal Fluid from Ugandans with Cryptococcal Meningitis and Immune Reconstitution Inflammatory Syndrome. *J Infect Dis*. 2014; 211:1597–606. This study highlights the evolution of the cellular response in the cerebrospinal fluid during primary cryptococcal meningitis and cryptococcal IRIS, showing activated intermediate monocyte trafficking during IRIS and distinct cellular changes in CSF compared to blood. *. [PubMed: 25492918]
29. Shoham S, Huang C, Chen JM, et al. Toll-like receptor 4 mediates intracellular signaling without TNF-alpha release in response to *Cryptococcus neoformans* polysaccharide capsule. *J Immunol*. 2001; 166:4620–6. [PubMed: 11254720]
30. Piccioni M, Monari C, Bevilacqua S, et al. A critical role for FcγRIIB in up-regulation of Fas ligand induced by a microbial polysaccharide. *Clin Exp Immun*. 2011; 165:190–201. [PubMed: 21605112]
31. Levitz SM, Nong S, Mansour MK, et al. Molecular characterization of a mannoprotein with homology to chitin deacetylases that stimulates T cell responses to *Cryptococcus neoformans*. *Proc Natl Acad Sci USA*. 2001; 98:10422–7. [PubMed: 11504924]
32. Koguchi Y, Kawakami K. Cryptococcal infection and Th1-Th2 cytokine balance. *Int Rev Immun*. 2002; 21:423–38.
33. Vecchiarelli A, Retini C, Pietrella D, et al. Downregulation by cryptococcal polysaccharide of tumor necrosis factor alpha and interleukin-1 beta secretion from human monocytes. *Infection and Immunity*. 1995; 63:2919–23. [PubMed: 7622213]
34. Chaka W, Heyderman R, Gangaidzo I, et al. Cytokine profiles in cerebrospinal fluid of human immunodeficiency virus-infected patients with cryptococcal meningitis: no leukocytosis despite high interleukin-8 levels. University of Zimbabwe Meningitis Group. *J Infect Dis*. 1997; 176:1633–6. [PubMed: 9395381]
35. Stenzel W, Muller U, Kohler G, et al. IL-4/IL-13-dependent alternative activation of macrophages but not microglial cells is associated with uncontrolled cerebral cryptococcosis. *Am J Path*. 2009; 174:486–96. [PubMed: 19147811]
36. Boulware DR, Bonham SC, Meya DB, et al. Paucity of initial cerebrospinal fluid inflammation in cryptococcal meningitis is associated with subsequent immune reconstitution inflammatory syndrome. *J Infect Dis*. 2010; 202:962–70. [PubMed: 20677939]
37. Dong ZM, Murphy JW. Intravascular cryptococcal culture filtrate (CneF) and its major component, glucuronoxylomannan, are potent inhibitors of leukocyte accumulation. *Infection and Immunity*. 1995; 63:770–8. [PubMed: 7868246]
38. Dong ZM, Jackson L, Murphy JW. Mechanisms for induction of L-selectin loss from T lymphocytes by a cryptococcal polysaccharide, glucuronoxylomannan. *Infection and Immunity*. 1999; 67:220–9. [PubMed: 9864219]
39. Kestens L, Seddiki N, Bohjanen PR. Immunopathogenesis of immune reconstitution disease in HIV patients responding to antiretroviral therapy. *Curr Opin HIV AIDS*. 2008; 3:419–24. [PubMed: 19373000]
40. DeSimone JA, Pomerantz RJ, Babinchak TJ. Inflammatory reactions in HIV-1-infected persons after initiation of highly active antiretroviral therapy. *Ann Int Med*. 2000; 133:447–54. [PubMed: 10975963]
41. Schacker TW, Nguyen PL, Beilman GJ, et al. Collagen deposition in HIV-1 infected lymphatic tissues and T cell homeostasis. *J Clin Inv*. 2002; 110:1133–9.

42. Annison L, Dompok A, Adu-Sarkodie Y. The immunological response of HIV-positive patients initiating HAART at the Komfo Anokye Teaching Hospital, Kumasi, Ghana. *Ghana Med J.* 2013; 47:164–70. [PubMed: 24669021]
43. Riou C, Tanko RF, Soares AP, et al. Restoration of CD4+ Responses to Copathogens in HIV-Infected Individuals on Antiretroviral Therapy Is Dependent on T Cell Memory Phenotype. *J Immunol.* 2015; 195:2273–81. The recovery of antigen-specific T cells is important in the immunopathogenesis of IRIS. This study describes the dynamics of antigen-specific CD4 T cells after ART initiation. *. [PubMed: 26195814]
44. Boulware DR, Meya DB, Bergemann TL, et al. Antiretroviral therapy down-regulates innate antiviral response genes in patients with AIDS in sub-saharan Africa. *J Acquir Immune Defic Syndr.* 2010; 55:428–38. [PubMed: 20838227]
45. Hasegawa A, Liu H, Ling B, et al. The level of monocyte turnover predicts disease progression in the macaque model of AIDS. *Blood.* 2009; 114:2917–25. [PubMed: 19383966]
46. Burdo TH, Lo J, Abbara S, et al. Soluble CD163, a novel marker of activated macrophages, is elevated and associated with noncalcified coronary plaque in HIV-infected patients. *J Infect Dis.* 2011; 204:1227–36. [PubMed: 21917896]
47. Lederman MM, Valdez H. Immune restoration with antiretroviral therapies: implications for clinical management. *JAMA.* 2000; 284:223–8. [PubMed: 10889597]
48. Shelburne SA 3rd, Hamill RJ, Rodriguez-Barradas MC, et al. Immune reconstitution inflammatory syndrome: emergence of a unique syndrome during highly active antiretroviral therapy. *Medicine (Baltimore).* 2002; 81:213–27. [PubMed: 11997718]
49. Murdoch DM, Venter WD, Feldman C, Van Rie A. Incidence and risk factors for the immune reconstitution inflammatory syndrome in HIV patients in South Africa: a prospective study. *AIDS.* 2008; 22:601–10. [PubMed: 18317001]
50. Achenbach CJ, Harrington RD, Dhanireddy S, et al. Paradoxical immune reconstitution inflammatory syndrome in HIV-infected patients treated with combination antiretroviral therapy after AIDS-defining opportunistic infection. *Clin Inf Dis.* 2012; 54:424–33.
51. Haddow LJ, Colebunders R, Meintjes G, et al. Cryptococcal immune reconstitution inflammatory syndrome in HIV-1-infected individuals: proposed clinical case definitions. *Lancet Inf Dis.* 2010; 10:791–802.
52. Wendel KA, Alwood KS, Gachuhi R, et al. Paradoxical worsening of tuberculosis in HIV-infected persons. *Chest.* 2001; 120:193–7. [PubMed: 11451837]
53. Cattelan AM, Trevenzoli M, Sasset L, et al. Multiple cerebral cryptococcomas associated with immune reconstitution in HIV-1 infection. *AIDS.* 2004; 18:349–51. [PubMed: 15075562]
54. Boelaert JR, Goddeeris KH, Vanopdenbosch LJ, Casselman JW. Relapsing meningitis caused by persistent cryptococcal antigens and immune reconstitution after the initiation of highly active antiretroviral therapy. *AIDS.* 2004; 18:1223–4. [PubMed: 15166545]
55. Katchanov J, Zimmermann U, Branding G, et al. Multiphasic and multifocal cryptococcal immune reconstitution inflammatory syndrome in an HIV-infected patient: interplay of infection and immunity. *Int J Inf Dis.* 2014; 18:87–9. Cryptococcal IRIS may not be limited only to the central nervous system, but may occur in different compartments. The case report highlights the organ-specificity and timing of cryptococcal IRIS after initiating antiretroviral therapy. *. [PubMed: 24161208]
56. Ingram PR, Howman R, Leahy MF, Dyer JR. Cryptococcal immune reconstitution inflammatory syndrome following alemtuzumab therapy. *Clin Inf Dis.* 2007; 44:e115–7.
57. Buggins AG, Mufti GJ, Salisbury J, et al. Peripheral blood but not tissue dendritic cells express CD52 and are depleted by treatment with alemtuzumab. *Blood.* 2002; 100:1715–20. [PubMed: 12176892]
58. Marie I, Guglielmino E. Non tuberculous anti-TNF associated opportunistic infections. *La Revue de medecine interne / fondee par la Societe nationale Francaise de Medecine Interne.* 2010; 31:353–60. [PubMed: 20381217]
59. Hage CA, Wood KL, Winer-Muram HT, et al. Pulmonary cryptococcosis after initiation of anti-tumor necrosis factor-alpha therapy. *Chest.* 2003; 124:2395–7. [PubMed: 14665529]

60. Wilson ML, Sewell LD, Mowad CM. Primary cutaneous Cryptococcosis during therapy with methotrexate and adalimumab. *Journal of drugs in dermatology : JDD*. 2008; 7:53–4. [PubMed: 18246698]
61. Jarvis JN, Casazza JP, Stone HH, et al. The phenotype of the *Cryptococcus*-specific CD4+ memory T-cell response is associated with disease severity and outcome in HIV-associated cryptococcal meningitis. *J Infect Dis*. 2013; 207:1817–28. [PubMed: 23493728]
62. Ishii K, Kawakami K. Up-to-date findings in the host defence mechanism to cryptococcus infection. *Medical mycology journal*. 2014; 55:J107–14. This review summarises our current understanding of the immune response to cryptococcal infection, creating the backdrop for understanding the immune responses observed during cryptococcal IRIS. *. [PubMed: 25231225]
63. Romani L. Immunity to fungal infections. *Nat Rev Imm*. 2011; 11:275–88.
64. Meya DB, Manabe YC, Castelnuevo B, et al. Cost-effectiveness of serum cryptococcal antigen screening to prevent deaths among HIV-infected persons with a CD4+ cell count < or = 100 cells/microL who start HIV therapy in resource-limited settings. *Clin Inf Dis*. 2010; 51:448–55.
65. Sungkanuparph S, Filler SG, Chetchotisakd P, et al. Cryptococcal immune reconstitution inflammatory syndrome after antiretroviral therapy in AIDS patients with cryptococcal meningitis: a prospective multicenter study. *Clin Inf Dis*. 2009; 49:931–4.
66. Grant PM, Komarow L, Andersen J, et al. Risk factor analyses for immune reconstitution inflammatory syndrome in a randomized study of early vs. deferred ART during an opportunistic infection. *PLoS One*. 2010; 5:e11416. [PubMed: 20617176]
67. Bisson GP, Molefi M, Bellamy S, et al. Early versus delayed antiretroviral therapy and cerebrospinal fluid fungal clearance in adults with HIV and cryptococcal meningitis. *Clin Inf Dis*. 2013; 56:1165–73.
68. Boulware DR, Meya DB, Muzoora C, et al. Timing of antiretroviral therapy after diagnosis of cryptococcal meningitis. *NEJM*. 2014; 370:2487–98. This multisite clinical trial sought to understand the effect of the timing of antiretroviral therapy following cryptococcal meningitis, however, the trial was discontinued before it could be powered to look at the effect of ART timing on the incidence of cryptococcal IRIS. *. [PubMed: 24963568]
69. Makadzange AT, Ndhlovu CE, Takarinda K, et al. Early versus delayed initiation of antiretroviral therapy for concurrent HIV infection and cryptococcal meningitis in sub-saharan Africa. *Clin Inf Dis*. 2010; 50:1532–8.
70. Zolopa A, Andersen J, Powderly W, et al. Early antiretroviral therapy reduces AIDS progression/death in individuals with acute opportunistic infections: a multicenter randomized strategy trial. *PLoS One*. 2009; 4:e5575. [PubMed: 19440326]
71. Jarvis JN, Meintjes G, Bicanic T, et al. Cerebrospinal Fluid Cytokine Profiles Predict Risk of Early Mortality and Immune Reconstitution Inflammatory Syndrome in HIV-Associated Cryptococcal Meningitis. *PLoS pathogens*. 2015; 11:e1004754. [PubMed: 25853653]
72. Stone SF, Price P, Keane NM, et al. Levels of IL-6 and soluble IL-6 receptor are increased in HIV patients with a history of immune restoration disease after HAART. *HIV Med*. 2002; 3:21–7. [PubMed: 12059947]
73. Meya, D.; Zziwa, G.; Okurut, S., et al. Monocyte Immune Responses in Cryptococcal Immune Reconstitution Inflammatory Syndrome. Conference on Retroviruses and Opportunistic Infections; February 22–26, 2015; Poster abstract number 815. This study demonstrates elevated intracellular cytokine expression by monocytes during cryptococcal IRIS. The study showed that IL-6 and TNF- α co-expression by monocytes was elevated in response to IFN- γ stimulation confirming that restored T cell signaling after initiating antiretroviral therapy was an important factor in the immunopathogenesis of cryptococcal IRIS. *
74. Coffey MJ, Phare SM, George S, et al. Granulocyte colony-stimulating factor administration to HIV-infected subjects augments reduced leukotriene synthesis and anticryptococcal activity in neutrophils. *J Clin Invest*. 1998; 102:663–70. [PubMed: 9710433]
75. Chau TT, Mai NH, Phu NH, et al. A prospective descriptive study of cryptococcal meningitis in HIV uninfected patients in Vietnam - high prevalence of *Cryptococcus neoformans* var *grubii* in the absence of underlying disease. *BMC Inf Dis*. 2010; 10:199.

76. Lee SJ, Choi HK, Son J, et al. Cryptococcal meningitis in patients with or without human immunodeficiency virus: experience in a tertiary hospital. *Yonsei Medical Journal*. 2011; 52:482–7. [PubMed: 21488192]
77. Lee YC, Wang JT, Sun HY, Chen YC. Comparisons of clinical features and mortality of cryptococcal meningitis between patients with and without human immunodeficiency virus infection. *Journal of Microbiology, Immunology, and Infection = Wei mian yu gan ran za zhi*. 2011; 44:338–45.
78. Mora DJ, Fortunato LR, Andrade-Silva LE, et al. Cytokine profiles at admission can be related to outcome in AIDS patients with cryptococcal meningitis. *PLoS One*. 2015; 10:e0120297. [PubMed: 25799044]
79. Naranbhai V, Chang CC, Durgiah R, et al. Compartmentalization of innate immune responses in the central nervous system during cryptococcal meningitis/HIV coinfection. *AIDS*. 2014; 28:657–66. This study points to the importance of the innate immune system in the localized response to *Cryptococcus* in the brain and describes the potential involvement of natural killer cells and monocytes, which could ultimately set the stage for the mechanisms leading to cryptococcal IRIS. *. [PubMed: 24451162]
80. Chang CC, Omarjee S, Lim A, et al. Chemokine levels and chemokine receptor expression in the blood and the cerebrospinal fluid of HIV-infected patients with cryptococcal meningitis and cryptococcosis-associated immune reconstitution inflammatory syndrome. *J Infect Dis*. 2013; 208:1604–12. This study highlights the compartmentalization of the immune response during cryptococcal IRIS showing that myeloid cell recruitment to the cerebrospinal fluid may predispose to cryptococcal IRIS. *. [PubMed: 23908492]
81. Wiesner DL, Boulware DR. Cryptococcus-Related Immune Reconstitution Inflammatory Syndrome(IRIS): Pathogenesis and Its Clinical Implications. *Curr Fungal Inf Rep*. 2011; 5:252–61.
82. Barber DL, Andrade BB, Sereti I, Sher A. Immune reconstitution inflammatory syndrome: the trouble with immunity when you had none. *Nat Rev Micro*. 2012; 10:150–6. This study although published outside the review period, points to an important hypothesis suggesting the role of innate and adaptive immune axes in the events leading up to the general mechanisms of IRIS. The authors suggest partial activation of monocytes in the absence of IFN- γ signaling from depleted T cells and *en masse* activation of these accumulated monocytes when ART restores the T cell signaling resulting in the immunopathology observed during IRIS. **.
83. Ziegler-Heitbrock L. The CD14+ CD16+ blood monocytes: their role in infection and inflammation. *J Leukoc Biol*. 2007; 81:584–92. [PubMed: 17135573]
84. Scriven JE, Rhein J, Hullsiek KH, et al. Early ART After Cryptococcal Meningitis Is Associated With Cerebrospinal Fluid Pleocytosis and Macrophage Activation in a Multisite Randomized Trial. *J Infect Dis*. 2015 This study highlights the possible deleterious role of macrophage activation when antiretroviral therapy was initiated within 7–11 days of the diagnosis of cryptococcal meningitis. This further stresses the role of monocyte recruitment into cerebrospinal fluid and the ensuing mortality which may occur without obvious symptoms to suggest overt IRIS. *.
85. Fazekas de St Groth B, Landay AL. Regulatory T cells in HIV infection: pathogenic or protective participants in the immune response? *AIDS*. 2008; 22:671–83. [PubMed: 18356596]
86. Tan DB, Yong YK, Tan HY, et al. Immunological profiles of immune restoration disease presenting as mycobacterial lymphadenitis and cryptococcal meningitis. *HIV Med*. 2008; 9:307–16. [PubMed: 18400078]
87. Seddiki N, Sasson SC, Santner-Nanan B, et al. Proliferation of weakly suppressive regulatory CD4+ T cells is associated with over-active CD4+ T-cell responses in HIV-positive patients with mycobacterial immune restoration disease. *Eur J Immun*. 2009; 39:391–403.
88. Meintjes G, Wilkinson KA, Rangaka MX, et al. Type 1 helper T cells and FoxP3-positive T cells in HIV-tuberculosis-associated immune reconstitution inflammatory syndrome. *Am J Resp & Crit Care Med*. 2008; 178:1083–9. [PubMed: 18755923]
89. Antonelli LR, Mahnke Y, Hodge JN, et al. Elevated frequencies of highly activated CD4+ T cells in HIV+ patients developing immune reconstitution inflammatory syndrome. *Blood*. 2010; 116:3818–27. [PubMed: 20660788]

90. Gupta M, Jafri K, Sharim R, et al. Immune reconstitution inflammatory syndrome associated with biologic therapy. *Current allergy and asthma reports*. 2015; 15:499. This review discusses the role of biologic therapies in the pathogenesis of immune reconstitution inflammatory syndrome and highlights the pros and cons of using these treatments. *. [PubMed: 25504263]
91. Tran HT, Van den Bergh R, Vu TN, et al. The role of monocytes in the development of Tuberculosis-associated Immune Reconstitution Inflammatory Syndrome. *Immunobiology*. 2014; 219:37–44. This study highlights the role of monocytes in the pathway leading up to immune reconstitution inflammatory syndrome associated with Tuberculosis. Due to the similarity in the primary immune responses to *Mycobacteria* and *Cryptococcus*, it is possible that there are similarities in the immunopathogenesis of IRIS involving both pathogens. *. [PubMed: 23958034]
92. Tran HT, Van den Bergh R, Loembe MM, et al. Modulation of the complement system in monocytes contributes to tuberculosis-associated immune reconstitution inflammatory syndrome. *AIDS*. 2013; 27:1725–34. [PubMed: 23807270]
93. Andrade BB, Singh A, Narendran G, et al. Mycobacterial Antigen Driven Activation of CD14+ +CD16- Monocytes Is a Predictor of Tuberculosis-Associated Immune Reconstitution Inflammatory Syndrome. *PLoS pathogens*. 2014; 10:e1004433. This study highlights the role of the classical monocytes in the pathogenesis of TB-IRIS. The functional role of the innate immune response in TB-IRIS was clearly proven in this study. The mechanisms reported here could be reflective of the immunopathogenesis of IRIS related to other opportunistic infections characterized by granulomatous inflammatory responses including Cryptococcal IRIS. **. [PubMed: 25275318]
94. Ho EL, Ronquillo R, Altmeyen H, et al. Cellular Composition of Cerebrospinal Fluid in HIV-1 Infected and Uninfected Subjects. *PLoS One*. 2013; 8:e66188. [PubMed: 23822975]
95. Scemla A, Gerber S, Duquesne A, et al. Dramatic improvement of severe cryptococcosis-induced immune reconstitution syndrome with adalimumab in a renal transplant recipient. *Am J Transp*. 2015; 15:560–4. The potential role of the TNF- α inhibitor as a therapeutic agent against cryptococcal IRIS is highlighted in this case report. The role of the inflammatory response mediated via TNF- α in cryptococcal IRIS is brought to light. *.
96. Sitapati AM, Kao CL, Cachay ER, et al. Treatment of HIV-related inflammatory cerebral cryptococcoma with adalimumab. *Clin Inf Dis*. 2010; 50:e7–10.
97. Hamilton SE, Wolkers MC, Schoenberger SP, Jameson SC. The generation of protective memory-like CD8+ T cells during homeostatic proliferation requires CD4+ T cells. *Nat Imm*. 2006; 7:475–81.
98. Jameson SC. T cell homeostasis: keeping useful T cells alive and live T cells useful. *Seminars in Immunology*. 2005; 17:231–7. [PubMed: 15826828]
99. Datta S, Sarvetnick N. Lymphocyte proliferation in immune-mediated diseases. *Trends in Immunology*. 2009; 30:430–8. [PubMed: 19699149]
100. Stone S, Price P, Keane N, et al. Levels of IL-6 and soluble IL-6 receptor are increased in HIV patients with a history of immune restoration disease after HAART. *HIV Med*. 2002; 3:21–37. [PubMed: 12059947]
101. Stone SF, Price P, Brochier J, French MA. Plasma bioavailable interleukin-6 is elevated in human immunodeficiency virus-infected patients who experience herpesvirus-associated immune restoration disease after start of highly active antiretroviral therapy. *J Infect Dis*. 2001; 184:1073–7. [PubMed: 11574925]
102. Tajima M, Wakita D, Noguchi D, et al. IL-6-dependent spontaneous proliferation is required for the induction of colitogenic IL-17-producing CD8+ T cells. *J Exp Med*. 2008; 205:1019–27. [PubMed: 18426983]
103. Jameson SC. Maintaining the norm: T-cell homeostasis. *Nat Rev Imm*. 2002; 2:547–56.
104. Martin-Blondel G, Delobel P, Blancher A, et al. Pathogenesis of the immune reconstitution inflammatory syndrome affecting the central nervous system in patients infected with HIV. *Brain*. 2011; 134:928–46. [PubMed: 21247931]
105. Vella AT, Dow S, Potter TA, et al. Cytokine-induced survival of activated T cells in vitro and in vivo. *Proc Natl Acad Sci USA*. 1998; 95:3810–5. [PubMed: 9520449]

106. Mahnke YD, Greenwald JH, DerSimonian R, et al. Selective expansion of polyfunctional pathogen-specific CD4(+) T cells in HIV-1-infected patients with immune reconstitution inflammatory syndrome. *Blood*. 2012; 119:3105–12. [PubMed: 22219223]
107. French M. Immune Reconstitution Inflammatory Syndrome: a reappraisal. *Clin Inf Dis*. 2009; 48:101– 7.
108. Grant PM, Komarow L, Lederman MM, et al. Elevated interleukin 8 and T-helper 1 and T-helper 17 cytokine levels prior to antiretroviral therapy in participants who developed immune reconstitution inflammatory syndrome during ACTG A5164. *J Infect Dis*. 2012; 206:1715–23. [PubMed: 23002445]
109. Rhein, J.; Hupler, HK.; Morawski, BM., et al. Adjunctive Sertraline for the Treatment of HIV-Associated Cryptococcal Meningitis. Conference on Retroviruses and Opportunistic Infections; February 22–26, 2015; Seattle, USA. p. Abstract 838This study highlights the potential role of Adjunctive Sertraline for treatment of Cryptococcal Meningitis and the preliminary observed decrease in incidence of Cryptococcal IRIS. *
110. World Health Organization. [Accessed April 1 2015] Rapid advice: Diagnosis, prevention and management of cryptococcal disease in HIV-infected adults, adolescents and children. Available at: http://www.who.int/hiv/pub/cryptococcal_disease2011

Key Points

- Poor cryptococcal antigen clearance is important in the development of cryptococcal IRIS.
- Monocyte recruitment into the CSF compartment during CM-IRIS may be essential for the subsequent cytokine-driven immunopathology that occurs during CM-IRIS.
- The confluence of residual cryptococcal antigen, chemokine-mediated trafficking of monocytes and ART-mediated CD4⁺ T cell redistribution into the CNS, may spawn exaggerated innate and adaptive immune responses that define cryptococcal IRIS.
- Future therapeutic interventions to prevent or treat cryptococcal IRIS could utilize our current understanding of the cellular kinetics of innate and adaptive cells in the CNS.

Figure 1. Model integrating Innate and Adaptive immune Axes in Cryptococcal IRIS
An integrated model showing events leading up to CM-IRIS. In the setting of high fungal burden, lack of effective type 1 T helper cell (Th1) response and/or a Th2 predominant response leads to poor cryptococcal clearance with residual cryptococcal antigen. Monocyte trafficking into the CSF occurs with increased expression of the chemokines CCL2 and CCL3.

In the absence of CD4⁺ T cells prior to ART initiation, the innate immune response consists mainly of primed macrophages/monocytes that take up antigen but do not have IFN- γ signaling from Th1 CD4⁺ T cells to become fully activated, thus antigen presenting cells accumulate and express suboptimal levels of innate signaling cytokines (e.g. IL-6). The confluence of factors including IL-6 from the myeloid cells and constant high-affinity MHC-peptide interactions resulting from partially cleared residual cryptococcal antigen in the setting of ART initiation could lead to lymphopenia-induced proliferation, expansion of CD4⁺ T cell memory cells and effective Th1 responses against residual cryptococcal antigen.

Occurring at the same time, IFN- γ signaling from the expanding CD4⁺ T cell population leads to the en masse activation of primed monocytes/macrophages leading to exaggerated innate cytokine responses. These combined responses lead to excessive inflammation, tissue destruction and within the closed CNS space, cryptococcal-IRIS involving the CNS can have morbid consequences.

Table 1

Characteristics of Studies related to Cryptococcal Immune Reconstitution Inflammatory Syndrome in Patients starting Antiretroviral Therapy.

Type of Study	Number of subjects	Baseline CD4/ μ L	Frequency of CM-IRIS (%)	Timing after ART start	CM-IRIS related mortality (%)	Country	Reference
Retrospective	10	39	50	2–11 months	..	USA	Jenny-Avital et al (2002) [11]
Prospective	434	86	2	4 weeks	67	South Africa	Lawn et al (2005)[12]
Retrospective	59	72	31	4 weeks	2	USA	Shelburne et al (2005) [13]
Retrospective	59	32	38	18 weeks	32	South Africa	Jenkin et al (2006) [14]
Retrospective	52	26	19	9.9 months	0	Thailand	Sungkanuparph et al (2007)[15]
Prospective	65	28	17	29 days	36	South Africa	Thana B et al (2009) [16]
Retrospective	120	13	8.3	8 months	30	France	Lortholary A et al (2005) [8]
Retrospective	154	..	17.5	27 days	11	China	Yan et al (2014) [17]
Prospective	44	20	16	12 weeks	40	Uganda	Kambugu et al (2010) [18]
Prospective	101	19	45	8.8 weeks	36	Uganda	Boulware et al (2010) [19]
Prospective	40	..	22.5	10 weeks	..	Brazil	Colombo et al (2011) [20]
Prospective	90	27	14	8.6 weeks	..	South Africa	Jarvis et al (2012) [21]
Prospective	13	16	25.5	..	19.8	South Africa	Chang et al (2013) [22]

Table 2**Risk Factors for Cryptococcal IRIS**

Microbiologic	Virologic	Immunologic	Therapeutic
Initial Fungal Burden - Higher fungal burden or cryptococcal antigen titer at CM diagnosis [13, 19, 21, 65]	Baseline and IRIS HIV RNA - Higher viral load prior to ART initiation - A robust virologic response to ART with a rapid decline in VL [16]	Baseline CD4 and Change with ART ±Lower pre-ART CD4 count [21, 66]. ±A robust immunologic responses to ART with rapid CD4 T cell increase [16] (excluding virologic failure).	ART Timing - Very early ART initiation in relation to diagnosis and therapy of CM [67–70]
Persistent Positive Cryptococcal CSF cultures. - Positive residual culture at the end of induction therapy when starting fluconazole 400mg [21, 22]		CSF WBC and protein - Poor initial CSF immune response as suggested by CSF white cell count (<5µL) cells/and protein (<50 mg/dl) when <i>Cryptococcus</i> disseminates to the CNS [36]	
		Cytokine responses - Low CSF IFN- γ , TNF- α , IL-2, IL-6, IL-8, and IL-17 [21, 22, 36]	

Abbreviations: ART- Antiretroviral therapy; CM- cryptococcal meningitis; WBC- white blood cells; CSF- cerebrospinal fluid; IL- Interleukin; IFN- Interferon; TNF- tumor necrosis factor; IRIS- immune reconstitution inflammatory syndrome.

Table 3

Summary of Paradoxical Cryptococcal-IRIS pathogenesis framework [19].

Phase	Immunologic Activity	Evidence in CM-IRIS Patients
Before ART	<ul style="list-style-type: none"> • Paucity of appropriate inflammation for cryptococcosis and/or • Inappropriate (Th2) responses resulting in: • Poor antigen clearance, pre-ART • Elevated CSF chemokine expression 	<ul style="list-style-type: none"> • ↓ TNF-α, G-CSF, GM-CSF, VEGF in serum • ↓ IFN-γ, G-CSF, TNF-α, IL-6 in CSF [21, 36] • ↑IL-4 pre-ART • Similar CSF CRAG at initial infection [36] • Higher CRAG pre-ART • ↑Expression of CCL2, CCL3 in CSF[71]
After Starting ART	<ul style="list-style-type: none"> • Increasing proinflammatory signaling from antigen presenting cells (APCs) due to persisting antigen burden and failure to clear antigen • Secondary activation of coagulation cascade 	<ul style="list-style-type: none"> • ↑ IL-6 from macrophages [72] then downstream • ↑ CRP production; ↑ IL-7 from APCs • ↑ D-dimer
At IRIS	<ul style="list-style-type: none"> • Effective response of innate and adaptive immune systems • Activation of coagulation cascade • Neuronal cell activation and damage • Aberrant innate cell trafficking 	<ul style="list-style-type: none"> • Th₁ ↑IFN-γ, VEGF; Th₁₇ ↑ IL-17 • Innate: ↑ IL-8, G-CSF, GM-CSF • ± Elevated CSF White blood cells • ± Elevated CSF Protein • Negative CSF culture • ↑ IL-6⁺, TNF-α⁺ monocytes[73*] • ↑ D-dimer • ↑ CSF FGF-2 • Trafficking of pro-inflammatory monocytes and CD4⁺ T cells into CSF [28].

Abbreviations: APC- Antigen presenting cell; IL- Interleukin; CRP- C reactive protein; IFN- Interferon; CSF- Cerebrospinal fluid; TNF- Tumor necrosis factor; CCL- C-C chemokine; ART- Antiretroviral Therapy; GM-CSF- Granulocyte macrophage colony stimulating factor; TH- T helper; CRAG- Cryptococcal Antigen; FGF-Fibroblast growth factor.

Table 4a

Profiles of Inflammation in Blood and CSF among patients with Cryptococcal Meningitis and Cryptococcal IRIS by HIV Serostatus.

Reference	Patient Description	CSF WBC/ μ L Median (range)	CSF Protein (mg/dL) Median (range)	CRAG Titer/Colony forming Units
Boulware, D et al, 2010 [36]	HIV Positive at CM-IRIS (n=33)	31 (5–85)	80 (57.5–122.5)	1:64 (CSF)
	HIV Positive at CM diagnosis (n=85)	5 (2.5–25)	40 (20–80)	1:1024 (CSF)
Bicanic, et al, 2009 [16]	HIV Positive at CM-IRIS (n=11)	23 (4–37)	72 (50–100)	1:256 (CSF)
	HIV Positive at CM diagnosis (n=65)	4 (0–85)	60 (40–130)	1:2048 (CSF)
Chang C et al, 2013 [22]	HIV Positive with CM-IRIS (baseline) (n=27)	10 (0–24)	70 (49–82)	1:1024 (Serum)
	HIV Positive without CM-IRIS (baseline) (n=63)	34 (8–144)	91 (61–154)	1: 1024 (Serum)
Shelburne, SA, et al, 2005 [13]	HIV Positive at CM-IRIS (n=18)	56 (27–93)	98 (54–202)	1:128 (CSF)
	HIV Positive without CM-IRIS (n=97)	12 (3–62)	91(46–141)	1:2048 (CSF)
Chau, T et al, 2010 [75]	HIV Negative (n=57)	287 (2–1080)	128 (35–263)	1:256 (Serum)
Lee SJ, et al, 2011 [76]	HIV Positive (n=11)	285 (1–286)	-	1: 256 (CSF)
	HIV Negative (n=9)	150 (6–150)	-	1:16 (CSF)
Lee YC, et al, 2011 [77]	HIV Positive (n=37)	26 (mean)	88	1:512 (77%) (Serum)
	HIV Negative (n=51)	86 (mean)	149	1:512 (50%) (Serum)
Mora, DJ, et al 2015 [78]	HIV Positive at CM diagnosis (n=30)	<20 (67%)	92 (61–119)	1:1024 (CSF)
	HIV Positive No CM (n=56)	<20 (67%)	34	-
	HIV Negative (n=48)	<20 (53%)	42	-
Jarvis et al 2012 [21]	HIV Positive with CM-IRIS (baseline) (n=9)	13	-	5.53 log ₁₀ CFUs
	HIV Positive without CM-IRIS (baseline) (n=9)	28	-	5.03 log ₁₀ CFUs

Comparison of different studies describing elements of the inflammatory response and antigen burden in CSF and serum for HIV negative patients with cryptococcal meningitis, HIV positive patients with and without IRIS. Percent in brackets indicates proportion of patients.

Table 4b

Cytokine/Chemokine Profiles in Blood and CSF among patients with Cryptococcal Meningitis and Cryptococcal IRIS by HIV Serostatus.

Reference	Patient Description	CSF Cytokines/Chemokines	Serum/Plasma Cytokines
Boulware et al, 2010 [19, 36]	HIV Positive at CM-IRIS (n=33)	↓IFN- γ , IL-6, IL-8, TNF- α at baseline ↑TNF- α , ↑IL-17, ↑VEGF, ↑IFN- γ at CM-IRIS event	↑IL-4, IL-17 at baseline ↓ TNF- α , G-CSF, GM-CSF, VEGF at baseline
	HIV Positive at CM diagnosis (n=85)	-	↓ G-CSF, ↓ IFN- γ , ↓ TNF- α , ↑MCP-1
Bicanic et al, 2009 [16]	HIV Positive at CM-IRIS (n=11)	-	↓IFN- γ , ↓TNF- α ↑IL-6 vs HIV Positive at CM diagnosis
	HIV Positive at CM diagnosis (n=65)	-	-
Chang et al, 2013 [22]	HIV Positive with CM-IRIS (baseline) (n=27)	↑CCL2, CCL3, CXCL10 in CSF compared to blood	-
	HIV Positive without CM-IRIS (baseline) (n=63)	-	-
Mora et al, 2015 [78]	HIV Positive at CM diagnosis (n=30)	↑IL-2, IL-4, IL-8, IL-10, IFN- γ , TNF- α , IL-17 compared to HIV negative ↑IL-2, IL-8, IL-17, IFN- γ , TNF- α vs HIV Positive no CM	↑IL-2, IL-8, IL-10, IFN- γ , TNF- α , IL-17 compared to HIV negative and HIV positive no CM
	HIV Positive No CM (n=56)	-	↑IL-4
	HIV Negative (n=48)	-	↑IL-12p40
Chaka et al, 1997 [34]	HIV Positive at CM diagnosis (n=16)	↑IL-1 β , IL-6, IL-8, IL-10 compared to controls without CM	-
Jarvis et al [71]	HIV Positive at CM diagnosis (n=90)	↑IL-6, IL-8, IFN- γ , CCL2, CCL3 ↓IL-2, IL-4, IL-12	-
Meya, et al [73*]	HIV Positive at CM-IRIS (n=11)		↑IL-6, TNF- α -producing monocytes compared to controls without CM-IRIS
Naranbhai et al, 2014 [79*]	HIV Positive at CM Diagnosis (n=23)	↑CXCR3, CX3CR1, CXCL10, ↓TNF- α in CSF compared to blood	-

Comparison of different studies describing cytokine profiles in CSF and serum for HIV negative patients with cryptococcal meningitis, HIV positive patients with and without IRIS. Abbreviations: IL- Interleukin; IFN- Interferon; TNF- tumor necrosis factor; IRIS- immune reconstitution inflammatory syndrome; VEGF- Vascular endothelial growth factor, G-CSF- granulocyte colony-stimulating factor; GM-CSF- granulocyte-macrophage colony-stimulating factor; CCL- chemokine (C-C motif) ligand; CXCL- chemokine (C-X-C motif) ligand.