

Fighting the Monster: Applying the Host Damage Framework to Human Central Nervous System Infections

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ABSTRACT The host damage–response framework states that microbial pathogenesis is a product of microbial virulence factors and collateral damage from host immune responses. Immune-mediated host damage is particularly important within the size-restricted central nervous system (CNS), where immune responses may exacerbate cerebral edema and neurological damage, leading to coma and death. In this review, we compare human host and therapeutic responses in representative nonviral generalized CNS infections that induce archetypal host damage responses: cryptococcal meningoencephalitis and tuberculous meningitis in HIV-infected and non-HIV-infected patients, pneumococcal meningitis, and cerebral malaria. Consideration of the underlying patterns of host responses provides critical insights into host damage and may suggest tailored adjunctive therapeutics to improve disease outcome.

In his book, *Beyond Good and Evil*, Friedrich Nietzsche once wrote, “Beware that, when fighting monsters, you yourself do not become a monster. . .” (1). In addition to the vagaries of human behavior, the phrase can also be applied to the dual effects of the human immune system—that a system highly evolved to protect the host from marauding pathogenic monsters can also be the instrument of its own destruction. The host damage–response framework captures this duality by asserting that microbial pathogenesis occurs along a continuum, wherein host damage can be a consequence of microbial virulence or the host immune response (2–4). In the setting of immunodeficiency, microbial virulence predominates, whereas during an “effective” immune response or after immune reconstitution, exuberant inflammation may contribute to excessive host damage. Within the spatial confines of the central nervous system (CNS), the host is particularly vulnerable to a robust host response that may lead to cerebral edema, restriction of blood flow, and hypoxia, with resultant brain damage, progressing to coma and possibly death (5).

Among studies of humans with CNS infectious diseases, recent studies have identified host cellular and cytokine/chemokine patterns compartmentalized within the CNS which are specific to the particular infectious disease syndrome. Consideration of patterns of pathogenesis may facilitate development of syndrome-specific clinical strategies that may help improve outcomes for these high-mortality conditions. Conversely, extrapolating therapeutics from one syndrome to another syndrome with different host responses may be unwise. In addition, patterns of immune responses to pathogens, heavily constrained by evolutionarily pressures, may give insight into autoimmune inflammatory disorders. For example, multiple sclerosis is one of the most common neurologic inflammatory disorders leading to permanent disability in young adults (6). Recently, similarities between the inflammatory response of non-HIV cryptococcal meningitis and the progressive form of multiple sclerosis led to validation of a cerebrospinal fluid (CSF) soluble CD27 (sCD27)-derived tissue inflammatory biomarker and helped to characterize this important form of multiple sclerosis as an inflammatory, rather than a neurodegenerative, disorder (7–9).

Pathogens associated with nonviral CNS infections are among the leading infectious causes of death worldwide. HIV, tuberculosis (TB), and malaria result in a large number of CNS infections, and along with bacterial meningitis due to respiratory pathogens such as *Streptococcus pneumoniae* they are among the top 10 causes of death (10). HIV/AIDS-related cryptococcal meningoencephalitis in sub-Saharan Africa is the leading regional cause of adult meningitis, with deaths nearing that attributable to tuberculosis in some studies (11). Viral meningitides are also a major source of infective mortality and have been the subject of a number of recent reviews (12, 13), but they will not be discussed here. In the present review, we summarize and compare the findings and impact of the host response on the pathophysiology and disease severity of these generalized CNS infections.

HIV-ASSOCIATED CRYPTOCOCCAL MENINGITIS AND IMMUNE RECONSTITUTION INFLAMMATORY SYNDROME (cIRIS)

Cryptococcus spp. cause a severe and often fatal meningoencephalitis in persons living with HIV/AIDS, accounting for 15 to 20% of AIDS-related deaths and resulting in approximately half a million deaths annually (11, 14, 15). The fungus is an encapsulated facultative intracellular pathogen that causes a deep tissue meningoencephalitis emanating from the meninges and the Virchow-Robin channels surrounding penetrating vessels within the brain parenchyma (16). This extensive tissue penetration beyond the superficial structures of the meninges potentially exposes the CNS immune system to a large fungal burden. In HIV/AIDS, susceptibility to *Cryptococcus* is a result of defects in adaptive immunity,

Published 26 January 2016

Citation Panackal AA, Williamson KC, van de Beek D, Boulware DR, Williamson PR. 2016. Fighting the monster: applying the host damage framework to human central nervous system infections. *mBio* 7(1):e01906-15. doi:10.1128/mBio.01906-15.

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TABLE 1 Host damage response to CNS infection syndromes

Syndrome	CSF cytokine/chemokine response pattern	Host damage evidence
Cryptococcal meningoencephalitis	HIV ⁺ patients: low IFN- γ , TNF- α HIV ⁻ patients: high IFN- γ , IL-6, and IL-10 but not TNF- α , IL-4, or IL-13	CSF sCD14 and sCD163, and histopathology CSF NFL
Pneumococcal meningitis	High IFN- γ and IL-2, complement components (e.g., C5)	CSF MMP-9, microglial NO and apoptosis-inducing factor
Tuberculous meningitis	High IFN- γ , IL-10, IL-13, VEGF	CSF cathelicidin LL-37
Cerebral malaria	Ang2, IL-8, IL-1RA, but not IFN- γ	Microvascular obstruction and endothelial cell activation
HIV ⁺ immune reconstitution inflammatory syndrome <i>Cryptococcus</i>	High IFN- γ , TNF- α , IL-6, G-CSF, VEGF, CCL11	Intermediate monocyte radical oxygen species
Tuberculous	High IFN- γ , CXCL10, IL12p40, IL-6, IL-17A	CSF MMP-2 and MMP-9, neutrophil-released S100A8/A9 (calprotectin) inducing apoptosis

centered around quantitative and qualitative T-cell defects (17). Residual immune activation is likely the result of antigen-specific and compensatory responses supported by interleukin-7 (IL-7) and IL-15 (18, 19), but it is insufficient to control the infection. Prior to treatment with antiretroviral therapy (ART), cryptococcal meningitis is characterized by high fungal burdens, suggesting a predominant role for fungal virulence. Pathogen virulence continues to play a role after institution of antifungal therapy, suggested by an inverse association between mortality and rates of CSF fungal clearance (20). In contrast, low levels of Th1-defining responses, such as gamma interferon (IFN- γ) production, as well as poorly expressed macrophage-associated tumor necrosis factor alpha (TNF- α) (Table 1) (21, 22), suggest minimal roles for immune-mediated damage in the ART-naive, HIV-infected host. A predominance of pathogen-mediated damage in ART-naive hosts may also explain lack of improvement with adjunctive potentially immune-suppressing corticosteroids that seeks to control host-mediated immune damage (23, 24).

On the other hand, treatment with ART leads to immune recovery, with cIRIS occurring in 15 to 30% of HIV-infected persons with cryptococcosis (25). This immune reconstitution syndrome is defined for CNS disease as a paradoxical clinical deterioration in the setting of negative fungal cultures or other explanations in a patient with previously diagnosed cryptococcal disease after initiation of ART (25). The recovering immune system encounters a large intracerebral fungal burden which persists despite antifungal therapy (26). Additionally, with antifungal therapy, fungicidal therapy releases intracellular and cell wall antigens for innate immune activation normally protected from immune surveillance in intact organisms by a thick immunotolerant cryptococcal capsule (27, 28). Prior to ART, viral and fungal antigens result in primed macrophages that have not received CD4⁺ T-cell help via IFN- γ signaling to become fully activated, thus resulting in a decoupling of the innate and adaptive immune systems (29, 30). The pre-ART lymphopenic environment is also believed to alter the function of the remaining CD4⁺ T cells, rendering them more pathogenic as the population expands after immune recovery (31). Supporting this are findings from a study by Chang et al. (32), who found that the risk of cIRIS was accentuated by low *Cryptococcus*-specific CD4⁺ lymphocyte Th1 IFN- γ responses prior to ART. However, data showing that higher CD8⁺ T cells predispose to cIRIS prior to ART suggest that the lymphopenic risk factor is specific to CD4⁺ T cells (33). After ART, expansion and activation of Th1 CD4⁺ sub-

sets drive macrophage activation and inflammatory cytokines with prominent IFN- γ and IL-6 production (Table 1). For example, Boulware et al. reported an elevated Th1-type response in cIRIS with a 2- to 3-fold elevated intrathecal IFN- γ , TNF- α , granulocyte colony-stimulating factor (G-CSF), vascular endothelial growth factor (VEGF), and eotaxin (CCL11) response but low CCL2 (34) and elevated serum inflammatory markers, such as IL-6 and C-reactive protein (35). Worsley et al. also reported a robust CSF IFN- γ response during cryptococcal IRIS (36). These findings with cIRIS are similar to the T-cell activation of IRIS associated with a range of other conditions, such as tuberculosis-associated immune reconstitution inflammatory syndrome (TB-IRIS), in which increased frequencies of effector memory, HLA-DR⁺, and Ki67⁺ CD4 cells and higher serum IFN- γ production are reported (37).

Increased recruitment of mononuclear immune cells to the intrathecal space has also been implicated in cIRIS, based on elevations in monocyte recruitment, growth factors, and chemokines such as G-CSF, VEGF, and CCL11 (22, 33, 34). These responses are echoed by IRIS related to other infections, such as pulmonary and disseminated tuberculosis, with increased frequencies of CD14⁺⁺ CD16⁻ "classical" monocytes in blood, associated with increased plasma levels of cytokines, including TNF- α (38). More recent data for cIRIS have described CD14⁺⁺ CD16⁻ classical subsets of monocytes in CSF samples at baseline developing into a more proinflammatory intermediate phenotype (CD14⁺⁺ CD16⁺) that produced radical oxygen species concurrent with cIRIS onset (39, 40). Predisposition to cIRIS is also associated with higher CSF ratios of monocyte-recruiting chemokines, such as CCL2::CXCL10 and CCL3::CXCL10 ratios, suggesting that more intact cell trafficking of partially activated macrophages prior to ART initiation may predispose to future cIRIS once CD4⁺ T-cell-mediated activation occurs (33). Early initiation of ART after treatment of cryptococcal disease results in higher levels of CSF macrophage cytokines, such as sCD14 and sCD163, suggesting macrophage activation with ART-mediated immune reconstitution (40). Macrophage activation in cIRIS thus acts in concert with T-cell-mediated damage and results in multiple, potentially damaging inflammatory effects.

In summary, these findings suggest that the coordinated response of a recovering immune system, confronting a large antigen load, results in activated T cells and monocyte axes (high Th1-M1 in synchrony), leading to inflammatory damage (Fig. 1,

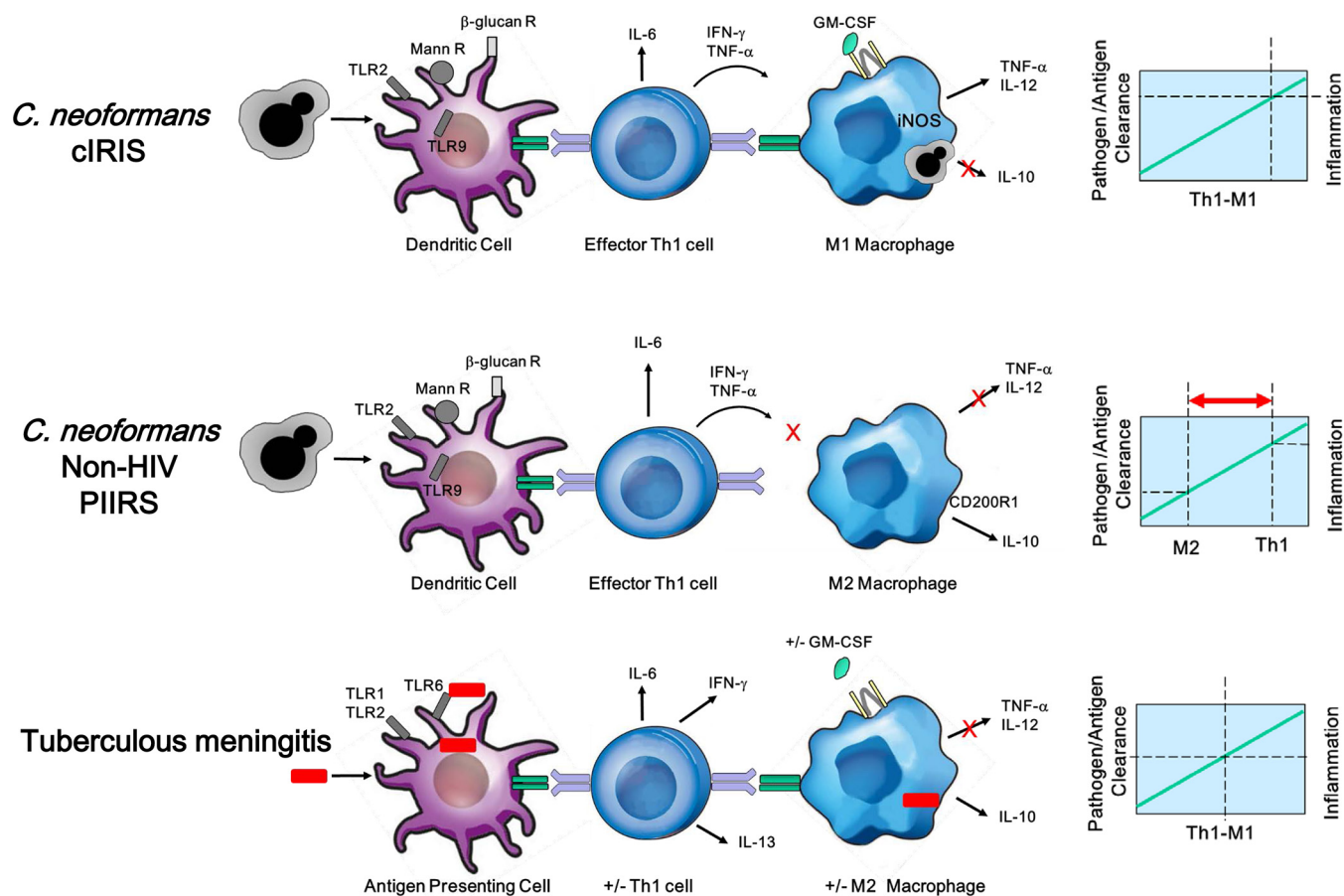


FIG 1 The host-damage framework applied to activation of the antigen-presenting cell–T-cell–macrophage activation pathways. (Left) Panels illustrate the predominant cellular response; (right) panels illustrate the potential contribution to host cell damage by the immune response. In the setting of cryptococcal meningoencephalitis, antifungal therapy followed by antiretroviral therapy results in pathogen activation of dendritic antigen-presenting cells through activation of TLRs, mannose receptors (Mann R), and β -glucan receptors (β -glucan R), resulting in a robust concordant Th1-M1 intrathecal response (cIRIS), whereas cryptococcal postinfectious inflammatory response syndrome (PIIRS) displays a discordant Th1-M2 CSF response (red arrow) with activated T cells causing increased inflammation but poor macrophage-mediated pathogen/antigen clearance. Tuberculous meningitis has an intermediary immunophenotype with moderately constrained inflammation and pathogen/antigen clearance.

top panel). Mechanisms of immune damage in the CNS are an area of active study and may include induction of cerebral edema (41), direct neurotoxic effects from macrophages (42), or metabolic programming of neurons by adjacent inflammatory signals (43). These findings also support the adjunctive role of immunosuppressive therapies, including corticosteroids, during cIRIS (high CSF IFN- γ state) (44), while immune-stimulating strategies, such as use of recombinant IFN- γ -1b, are most likely to be useful for microbiologically refractory disease in HIV-naïve individuals (especially those with low CSF IFN- γ levels), while it is likely to exacerbate cIRIS (45, 46).

CRYPTOCOCCAL MENINGOENCEPHALITIS IN PREVIOUSLY HEALTHY ADULTS AND POSTINFECTIONOUS INFLAMMATORY RESPONSE SYNDROME (PIIRS)

Although HIV-related cryptococcal disease is declining in high-income countries due to ART access, non-HIV-related cryptococcosis represents an ever-increasing proportion of cases (47). Two general categories of non-HIV patients exist. The first are those with preexisting conditions, such as Cushing's syndrome or immune suppression by immunotherapy, cancer chemotherapy, or

transplant conditioning. A second category of disease afflicts previously healthy, apparently immunocompetent individuals (15). Previous clinical research in non-HIV disease emphasized microbiological clearance of live organisms as a key to the resolution of pathophysiology, similar to ART-naïve cryptococcosis, with cerebrospinal fluid culture negativity at 2 weeks an important prognostic marker (48). In addition, an assumption of T-cell defects based on HIV-related susceptibilities has prompted recommendations for Th1-biasing immune therapy, such as IFN- γ for refractory cases among non-HIV-infected patients (44). However, reconsideration of the pathophysiology in these patients has occurred recently, with greater attention to a role for the host damage response. For example, in transplant-related cryptococcosis, clinical failure has been related to adjunctive reductions in immune suppression, potentiating an IRIS-like syndrome (49). Such reductions in immune suppression are commonly undertaken in immunosuppressed hosts during therapy to potentiate the immune response against pathogens. Yet, augmenting the immune response in patients with CNS infection may not be beneficial considering the damage-response framework. In contrast, other investigators have suggested a role for corticosteroids in non-

HIV-related cryptococcosis (50). In addition, some previously healthy individuals with *Cryptococcus* infection have appeared to have defective macrophage signaling, suggested by STAT5-blocking antibodies to GM-CSF, with retention of normal T-cell activity (51, 52). Similarly, G-CSF given to lymphopenic, HIV-uninfected hosts resulted in unmasking of clinical symptoms of infection (53). Thus, mechanisms related to immune-mediated host damage in this population have remained unclear.

To help clarify these issues, a prospective immune analysis of previously healthy patients with active CNS cryptococcosis was recently conducted (54). Previously healthy patients were chosen, both because they represent an important subpopulation of susceptible hosts and also because this provided greater patient sample uniformity due to a lack of confounding by variable levels of immunosuppression present in other at-risk populations, such as solid organ transplant patients or those receiving corticosteroids. All patients were severely ill with severe mental status changes despite antifungal therapy and their apparent immunocompetent state. A majority required ventricular-peritoneal shunting to relieve CSF obstruction from choroidal inflammation. Surprisingly, all had negative CSF cultures after standard courses of antifungal therapy, suggesting clinical deterioration despite effective microbiological control of their disease. This suggested that, unlike the ART-naïve HIV⁺ cryptococcus-infected patients, pathogen virulence was not the predominant cause of their refractory illness. However, much like cIRIS, the patients with CNS disease exhibited a robust intrathecal cellular Th1 response, with both CD4⁺ and CD8⁺ activation, as measured by HLA-DR positivity with production of high levels of Th1-biasing IFN- γ and inflammatory cytokines, such as IL-6, and a relative lack of Th2 cytokines, such as IL-4 and IL-13 (Table 1). Activation of T cells was also suggested by *ex vivo* study results, which demonstrated high levels of both CD4 and CD8 production of IFN- γ when T cells from CSF samples were cocultured with cryptococcal antigen-exposed dendritic cells. In addition, specific biomarkers of tissue inflammation, such as sCD27/T-cell ratios, suggested that inflammation was not restricted to the CSF alone but was also present within the meninges or brain parenchyma (8). Analysis of brain biopsy specimens and of a second set of autopsy specimens confirmed extensive meningeal and Virchow-Robin channel macrophage and T-cell infiltration, suggesting an immune etiology for the cerebral edema accompanying non-HIV-related cryptococcosis (9). This inflammatory response was also accompanied by elevated levels of CSF neurofilament light chain (NFL), a marker of axonal damage (55), suggesting ongoing host neurological damage. This cryptococcal postinfectious inflammatory response syndrome (PIIRS, pronounced “pēers”, or cPIIRS), associated with host cell damage, was present whether the original infecting organism was *Cryptococcus neoformans* or *Cryptococcus gattii*, the latter of which is typically believed to cause greater inflammation (56). However, both among persons with anti-granulocyte-macrophage colony-stimulating factor (GM-CSF) autoantibody and those without, tissue macrophage recruitment to the site of CNS infection was intact but brain histopathology demonstrated an M2 macrophage polarization (CD68⁺ CD200R1⁺) and poor phagocytosis of fungal cells, as identified by calcafluor white staining (54). This finding was supported by significantly high IL-10 and low TNF- α levels intrathecally (the latter predominantly produced by tissue macrophages). IL-10 production by alternatively activated M2 macrophages has been associated with other diseases for which

there is poor microbial/antigen clearance, such as lepromatous leprosy (57). The known plasticity of monocytes coupled with this apparent Th1-M2 discordance suggests that those non-HIV-infected patients with severe cryptococcal meningoencephalitis may have a downstream monocyte defect in the efferent arm of the immune response (Fig. 1, second panel). This Th1-M2 dissociation in cPIIRS thus results in a damaging T-cell host response but poor antigen clearance by macrophages, resulting in a prolonged clinical course of 1 to 2 years in many of the severe cases. Clearly, with up to 30% mortality in non-HIV-related cryptococcosis, clinical identification of patients with cPIIRS is essential to rational therapy. Novel approaches taking into account immune-mediated host damage may reduce mortality in these refractory clinical cases.

TUBERCULOUS MENINGITIS

According to the World Health Organization (WHO), tuberculosis affects 1/3 of the world’s population, with 1.5 million deaths annually; it is the second leading infectious cause of death after HIV (58). Although tuberculous meningitis (TBM) occurs in approximately 1% of tuberculosis cases, it is most frequent and severe in children (59), with an estimated mortality rate of 15 to 75% (60) and adverse sequelae in 10 to 85% of patients (61).

Steroid responsiveness in TBM suggests that immune-mediated host damage may play a significant role in disease pathology (62–64). Further immune studies of TBM in children suggested an intermediately activated T-lymphocyte/monocyte immunophenotype (Fig. 1, third panel). It is characterized by intrathecal Th1 markers, such as IFN- γ , accompanied by Th2 and M2 markers, such as IL-13 and IL-10, respectively, suggesting a mixed T-cell and macrophage polarity (Table 1) (65). In addition, elevated levels of cathelicidin LL-37, VEGF, and CCR5 suggest intact macrophage recruitment but deficient TNF- α expression, indicating a lack of effective macrophage activation, similar to cryptococcal PIIRS (66). Cathelicidin LL-37 is an antimicrobial peptide found in the lysosomes of macrophages and neutrophils and is important in the vitamin D receptor pathway. Indeed, vitamin D deficiency has been associated with tuberculosis progression (67), but vitamin D has not been associated with other CNS infections, such as cryptococcosis (68).

With such a robust CSF IFN- γ response in TBM, similar to that in cIRIS and cPIIRS, it is interesting that the adjunctive corticosteroid dexamethasone reduced mortality by 31% through 9 months (64). Corticosteroids, however, did not reduce chemokine expression in TBM (69), suggesting that clinical effectiveness could be more related to control of associated noninflammatory parameters, such as cerebral edema, which is the primary target of moderate doses of corticosteroids (1 to 1.5 mg/kg of body weight of prednisone equivalent) used in these studies (70). In contrast, high doses of corticosteroids of 18 mg/kg/day are typically used for severe inflammatory states, such as cerebral vasculitides (71). Indeed, alternative pathways of therapy are indicated by recent data suggesting that corticosteroids at achievable doses may reduce cerebral edema by coordinate regulation of angiopoietin-1 and VEGF, which are direct modulators of vasogenic brain edema and the blood-brain barrier, independent of inflammation (72). Thus, excess, damaging inflammation may fulfill the role of a prognostic marker of poor outcome because of its association with cerebral edema, but control of inflammation apart from cerebral edema may or may not affect outcome. This concept of the independence

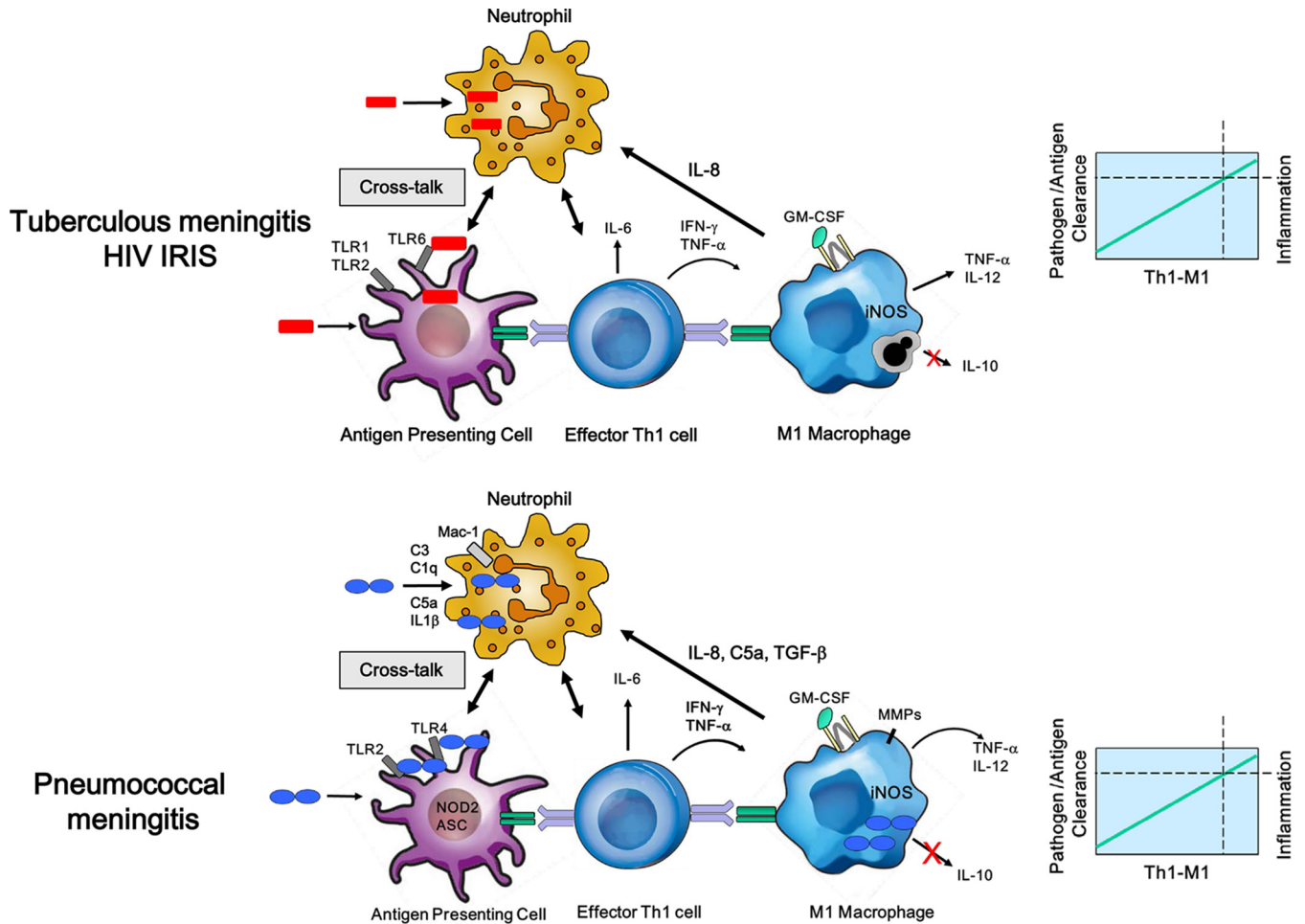


FIG 2 Host-damage framework in diseases with an additional CSF neutrophilic response. In IRIS related to tuberculous meningitis and pneumococcal meningitis, activation of antigen-presenting cells through TLRs leads to T-cell activation and cytokine release (IL-6, IFN- γ , TNF- α , GM-CSF) and a macrophage response leading to further TNF- α and IL-12 production as well as cross talk with neutrophils. Further activation of neutrophils follows pathogen phagocytosis and, in pneumococcal meningitis, to complement and IL-1 β activation. In both cases, the addition of neutrophil activation leads to additional pathogen clearance after therapy but at the cost of host-damaging intrathecal inflammation.

of a prognostic marker from a treatment surrogate is an important therapeutic principle that, if ignored, may lead to a false linkage of pathophysiology to treatment expectations (73, 74). The failure of a recent drug trial based on a mistaken belief that a microbiological prognostic marker of *Mycobacterium* spp. pathogen clearance would provide a good treatment surrogate is a good demonstration of this concept. That study hypothesized that increasing clearance by using a higher dose of antibiotic would lead to better outcomes. However, although pathogen clearance was increased, there were more deaths because of untoward effects of the higher dose of antibiotic (75). Currently, in cases of neurological infections, higher doses of corticosteroids or additional immunosuppressants have been proposed as adjunctive therapy in TBM, to reduce both innate and adaptive immune responses through the extracellular signal-regulated kinase 1/2 and NF- κ B pathways (76). However, the threat of adjunctive immunosuppressive therapies to effective microbiological control is a clear danger in diseases such as TBM and cryptococcosis and could result in adverse outcomes. Clearly, precise immunophenotyping analyses, to stratify patients and monitor therapeutic interventions, will be required for rational design and selection of adjunctive therapies.

TBM IRIS IN HIV

Because of the scale-up in ART and large numbers of TB-infected individuals globally, TB-related immune reconstitution inflammatory syndrome (TB-IRIS) after ART initiation is a significant contributor to the health care burden, especially in high-TB-HIV coinfection incidence populations (77). CNS involvement is the most severe form of TB-IRIS with a high associated mortality (78). Interestingly, in addition to a monocytic infiltration typical of TBM, TBM-IRIS results in an additional neutrophilic CSF infiltration (79), distinguishing it from non-HIV-associated TBM in children and cIRIS in adults (80) (Fig. 2, top panel). Similar to cIRIS, TBM-IRIS has been associated with elevations in Th1 cytokines, such as IFN- γ , and a delayed-type hypersensitivity response (81) (Table 1). More recently, Marais et al. studied compartmentalized CSF immune responses from HIV TBM patients at TBM diagnosis, start of ART, and at IRIS diagnosis. TBM-IRIS was associated with elevated Th1 markers (IFN- γ , CXCL10) and inflammatory markers such as IL-6, similar to that in cIRIS, with the addition of neutrophil recruitment markers S100A8/A9 (calprotectin), matrix metalloproteinase 9 (MMP-9), and its inhibitor,

tissue inhibitor of metalloproteinases 1 (80). Production of IL-8 by endothelial cells and macrophages is important for neutrophil influx in pulmonary TB (82), and IL-8 is also elevated in TBM (83), although IL-8 has not been studied specifically in TBM-IRIS. In addition, based on a murine TB model showing that IL-17A-induced S100A8/A9 was key in neutrophil accumulation and lung infiltration (84), CSF IL-17A levels were measured and found to increase with development of TBM-IRIS, supporting previous suggestions that IL-17 may be important in host cell-mediated immune damage (85). A predilection for TBM-IRIS is also related to preceding high CSF IFN- γ and TNF- α levels with neutrophilia, in contrast to the lymphopenia and low CSF IFN- γ levels predictive of cIRIS (79). However, in TB as in cIRIS, pathogen antigen load is a major driver of this paradoxical pathological immune response process.

Corticosteroids have been shown to be useful in TB-IRIS in a randomized, placebo-controlled trial, although patients with CNS involvement were excluded (86), but symptomatic improvement in TBM-IRIS has been reported with steroid use (80). However, CSF inflammation (e.g., IFN- γ but not TNF- α changes) persisted following ART in TB-IRIS, despite adjunctive corticosteroids, again suggesting that moderate doses of corticosteroids may act primarily by control of cerebral edema, similar to the experience in TBM (69, 80). However, effects on neutrophil markers such as MMP-9 have been variable, with some investigators showing decreases with steroid treatment (87) and others not showing reductions (80). These findings have also suggested to some that immunomodulatory treatment options more potent and specific than corticosteroids need to be explored for the prevention and management of TBM-IRIS (80), but the risks again must be balanced, as described above.

PNEUMOCOCCAL MENINGITIS

Pneumococcal infection causes approximately 2 million deaths and requires medical outlays of hundreds of billions of dollars per year (88). *Streptococcus pneumoniae* frequently colonizes the nasopharynx but can spread from the airway to the lower respiratory track, sinuses, middle ears, or to the CNS. T helper cell (Th-17) T-cell responses are key to controlling colonizing bacteria (89), and they are mediated by recruitment of macrophages in naive hosts and of neutrophils in previously exposed individuals. The organism is the most frequent bacterial cause of meningitis in adults (excluding Africa) and can lead to host damage through a variety of mechanisms, resulting in the highest case fatalities and neurological disability rates of the bacterial meningitides (90, 91). The virulence factor/toxin pneumolysin plays a pivotal role in both direct host damage and immune recognition/inflammation (92). Pneumolysin has the capacity to form membrane pores to lyse host cells, but it also activates innate immunity, as it stimulates caspase-1-dependent processing of IL-1 β , dependent on the nucleotide-binding oligomerization domain (NOD)-like receptor P3 inflammasome (93). Adaptive immunity is also stimulated by pneumolysin, which is recognized by Toll-like receptor 4 (TLR4) on antigen-presenting cells, which, in turn, induces production of inflammatory cytokines such as IFN- γ and IL-17A by T cells (94). Interestingly, TLR4-deficient mice did not differ from wild-type mice in their host response, while TLR2/4 double deficient mice showed a marked reduction in inflammatory mediators and improved outcome compared to wild-type mice or those with single TLR deficiency with pneumococcal meningitis, suggesting a role

for TLR signaling in CNS-related host damage (95). In a study of 28 patients with pneumococcal and meningococcal meningitis, CSF IFN- γ levels were significantly higher and borderline higher IL-2 levels were observed in pneumococcal compared with meningococcal patients across time, implying a greater Th1 bias for pneumococcal meningitis (Table 1). In another study of 45 patients, those with pneumococcal meningitis had significantly higher CSF levels of IFN- γ , CCL2 (MCP-1), and MMP-9 than those with meningococcal or *Haemophilus influenzae* meningitis (96). High MMP-9 expression has been associated with blood-brain barrier damage and neurologic sequelae (97). *In vitro* coculture of astrocytes and pneumococcal cell walls with microglia also led to these resident brain macrophages producing nitric oxide and causing neuronal toxicity, which was suppressed by dexamethasone (98); microglia are key in the recruitment of effector immune cells from the periphery following infection and may undergo a caspase-induced apoptosis leading to cytokine release in response to pneumococcus (99). Activation of a strong T-cell response in bacterial meningitis thus has some features common to TBM and *Cryptococcus* infection of HIV-infected and uninfected individuals. However, in bacterial meningitis, robust recruitment of neutrophils to the intrathecal space is a prominent feature, as in TBM-IRIS (Fig. 2, bottom panel), and requires the β 2 integrin Mac-1, resulting in the generation of neutrophil serine proteases cathepsin G and neutrophil elastase (100–102). Despite the key role of neutrophils in controlling bacteria, they also contribute to host cell damage, similar to TBM-IRIS. The production of NADPH oxidase-dependent reactive oxygen species contributes to collateral host cell damage in tissues (103) but, paradoxically, is not required to kill *S. pneumoniae*. In the CNS, neutrophil-produced myeloid-related protein 14 (MRP14) was found in a mouse model to exacerbate meningeal inflammation even after treatment with antibiotics in a TLR4-, CXCL2-dependent manner, again implicating TLR signaling in CNS host damage (104). Interestingly, treatment with the MRP14 antagonist paquinimod reduced inflammation and disease severity in mice, suggesting that identification of key host damage pathways may result in effective adjunctive therapies (104). In addition, moderate-dose adjunctive corticosteroids (e.g., dexamethasone equal to prednisone at 1 mg/kg four times daily for 4 days) has been used successfully in bacterial meningitis with significantly decreased mortality in human clinical trials (105, 106).

A particularly exciting development is the finding that adjunctive treatment with anti-complement C5 antibodies reduced mortality in pneumococcal meningitis in humans (107). Recently, an additive effect of dexamethasone and anti-C5 antibodies as adjunctive treatment has been shown in experimental pneumococcal meningitis (108). This provides a strong precedent that addition of specific inflammatory inhibitors in neuroinflammatory infections may potentiate the effects of corticosteroids on cerebral edema. Complement components are expressed in the CSF by microglia as well as injured astrocytes and neurons in response to inflammatory cytokines (109, 110). The complement cascade is activated through classical and alternative pathways after specific pathogen-cell interactions, although *S. pneumoniae* expresses several anticomplement strategies, such as pneumococcal surface protein C (PspC), which binds human factor H and blocks C3 convertase. Together, PspA and PspC proteins limit complement-mediated adherence (111). However, proinflammatory cytokines such as IL-6 have also been demonstrated to upregulate expres-

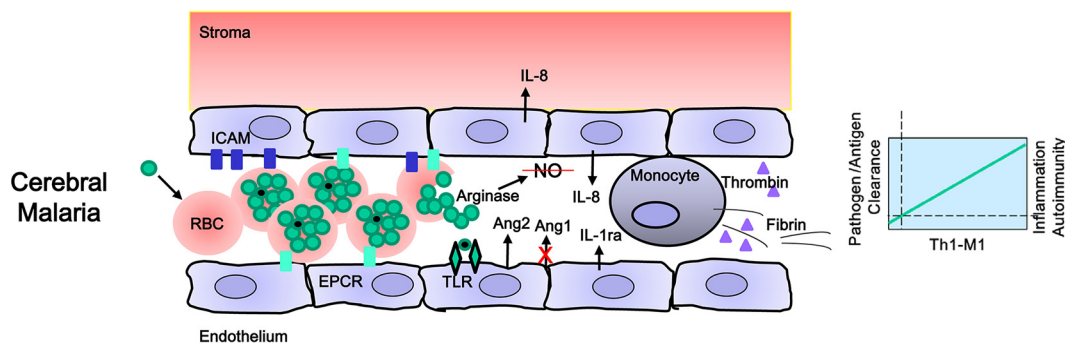


FIG 3 Host-damage framework applied to cerebral malaria with a predominance of endothelial injury and limited inflammation. Parasite infection of RBCs results in binding to the endothelial receptors ICAM and EPCR, followed by microvascular sequestration. Released parasite products (black dot) and RBC arginase activate TLRs and lead to NO inhibition. Activated endothelial cells release intravascular IL-8 and IL-1RA, leading to a monocyte inflammatory response that results in thrombin and fibrin production, potentiating endothelial injury and sequestration.

sion of C5aR in both liver and lung tissue, and anti-IL-6 antibody reduces complement activation during sepsis (112). Newly described roles for inflammatory pathways in complement activation, independent of the organism, may thus suggest studies of complement in inflammatory syndromes whose pathogens typically do not activate complement strongly, such as infection with encapsulated *Cryptococcus* (113).

CEREBRAL MALARIA

In countries where malaria is endemic, cerebral malaria caused by *Plasmodium falciparum* affects primarily children and malaria-naïve visitors, with case fatality proportions of 15 to 25% (114). The pathogenesis of cerebral malaria is incompletely understood, but it is distinguished from the infections described above by a predominance of endothelial activation by parasites sequestered in the brain microvasculature accompanied by only a modest inflammatory response (Fig. 3) (115). Pathological studies of individuals dying of the disease demonstrate intravascular parasites, vascular congestion and obstruction (116–119), and endothelial cell activation (120). In children this has been associated with a breakdown of the blood-brain barrier, ring hemorrhages, and cerebral edema, as evidenced by increased brain weight (121). In a recent study of 168 African children with cerebral malaria, 84% of the 25 children that died had brain swelling/volume findings on magnetic resonance imaging (MRI), whereas only 27% of the survivors had such findings, which eventually improved (122). An important confounder in the MRI study was that over 80% of the cerebral malaria patients had a history of seizures.

Notably absent are reports of autopsy samples with lymphocytes or neutrophils localizing to parenchymal areas of parasite sequestration or the CSF (116, 118, 121). In contrast, intravascular monocytes, fibrin, and platelets have been reported to localize with sequestered parasites in children, particularly those who are HIV⁺ (111, 123). Thus, a lack of a marked leukocyte infiltration has focused attention on endothelial cells and the local vascular environment as mediators of the pathology that leads to impaired consciousness and provides a sharp contrast to the other CNS infections we have described.

Cerebral edema associated with cerebral malaria is also unusual in that it was not found to be responsive to steroids in two randomized controlled trials (119, 124). In addition, cerebral edema may differ in children from that of adults, as brain weights in adults have been reported not to increase and expression of the

brain water channel, aquaporin 4 (or VEGF), did not show elevations in adult patients dying of cerebral malaria (125, 126). A significant increase in endothelial regulator angiopoietin 2 (Ang2) and a decrease in angiopoietin 1 (Ang1) have been reported in both adult and pediatric cerebral malaria patients (127–130); Ang2 antagonizes Ang1, resulting in an increase in vascular permeability, NF- κ B activation, and endothelial receptor upregulation (131). It is thus possible that high Ang2/Ang1 ratios could have a greater impact in children (125). In addition, prolonged seizures, more typical of cerebral malaria in children, may contribute to cerebral edema and abnormal MRI findings, suggesting cerebral malaria (122, 126). The specific induction stimulus for Ang2 is unknown but could include hypoxia, thrombin, and low nitric oxide (NO) levels, which have been associated with areas of parasite sequestration (132–134). A role for Ang2 is also consistent with the lack of steroid responsiveness, as *in vitro* studies have shown that dexamethasone does not decrease Ang2 expression in human brain microvascular endothelial cells (72). Clearly, much remains to be understood regarding the pathogenesis of cerebral malaria and the impact of possible edematous states.

Parasite cytoadherence to endothelial cell surface receptors is thought to be mediated primarily by a *P. falciparum*-specific, polymorphic, parasite-produced protein, PfEMP1, that is exported to the red blood cell (RBC) surface (135). In the brain microvasculature, intercellular adhesion molecule 1 (ICAM-1), which binds to a subset of PfEMP1 molecules, has been found upregulated in areas of parasite sequestration (118, 132). Recently, expression of group A PfEMP1s that bind to the endothelial protein C receptor (EPCR) (136) have been associated with severe malaria (137–139). Intriguingly, EPCR expression levels have been reported to be reduced in the microvasculature, with sequestered parasites and soluble EPCR levels increased in the CSF of cerebral malaria patients, suggesting that parasite binding stimulates receptor shedding (132). The effects of PfEMP1/EPCR or ICAM-1 binding are still under investigation, but it has been proposed that PfEMP1 binding to EPCR disrupts the production of activated protein C, inhibiting its anticoagulant and cytoprotective effects (132, 140). The low arginine and NO levels associated with cerebral malaria (141–143) as well as the ability of parasite material released during infected RBC rupture to stimulate TLRs (144) could also contribute to local endothelial activation, vascular obstruction, and local areas of hypoxia (133, 140). Increasing NO bioavailability as an

adjunct to antimalaria chemotherapy has been tested in several recent clinical trials (145). One trial found the administration of L-arginine to adult severe malaria patients in addition to intravenous artesunate to be safe, but there was no alteration in endothelial NO bioavailability as measured by reactive hyperemia peripheral arterial tonometry (146). The plasma arginine levels achieved were lower than predicted and could have contributed to the lack of efficacy. Direct NO inhalation has also been tested in conjunction with antimalaria chemotherapy in pediatric cerebral malaria patients. Again, the intervention was safe but did not significantly alter morbidity or mortality (147), suggesting that alternative strategies are needed to improve treatment of cerebral malaria.

The association of specific cytokines with cerebral malaria has been more difficult to define, as results vary between groups, possibly due to underlying coinfections or individual genetic variation (115). Several groups have found both IL-8 and interleukin 1 receptor antagonist (IL-1RA) to be significantly increased in CSF samples from patients with cerebral malaria compared to samples from those with severe malaria (Table 1) (148, 149). Interestingly, both these cytokines can be produced by endothelial cells (150, 151), and the finding that IL-8 levels are higher in CSF than in serum during cerebral malaria is also consistent with production in the CNS (149). *In vitro*, the incubation of *P. falciparum*-infected RBCs with human brain microvascular endothelial cells has also been shown to activate the NF- κ B pathway, leading to the production of a number of chemokines, including IL-8 (152). It is possible that IL-8 plays a role in recruiting the monocytes observed in pediatric patients, as well as enhancing angiogenesis (151), while IL-1RA could act to downregulate the inflammation response by directly competing with IL-1 α and IL-1 α receptor stimulation (153). Together, the data to date suggest that parasite sequestration in the brain microvasculature coupled with the subsequent rupture of infected RBCs during parasite release leads to marked endothelial cell activation and vascular obstruction with minimal lymphocyte infiltration or activation in the CNS. The resulting neuronal damage is likely due to hypoxia and metabolic disruption and, in children, is exacerbated by cerebral edema and seizures.

CONCLUSIONS

In summary, the host damage-response framework is exemplified in the generalized CNS infectious syndromes considered here. Defined repertoires of host cytokine/chemokine immune response profiles in these syndromes lead to a common endpoint of host neuronal damage. An interesting finding from our review is that neuroinfections characterized by significantly high CSF IFN- γ and T-cell inflammation may be more likely to benefit from adjunctive corticosteroid use, i.e., cryptococcal IRIS and PIIRS, pneumococcal meningitis, TBM, and TBM-related IRIS, versus cerebral malaria, where parenchymal and intrathecal inflammation is minimal. However, with regard to treatment, CNS inflammation may only be a marker of other pathologies, such as cerebral edema, since moderate doses of corticosteroids that are clinically beneficial have little effect on intrathecal inflammation in diseases such as TBM and TB-IRIS. However, caution must be exercised in the selection of immune-modulating therapies, as exemplified by the finding that adjunctive immune stimulators such as recombinant IFN- γ 1b benefited only small subsets of patients who had low CSF IFN- γ levels and poor microbiological control in the case of ART-naive HIV-related cryptococcal disease

(45, 46) and could be detrimental in inflammatory states such as PIIRS, where a high-IFN- γ inflammatory state is mistaken for refractory disease (54). Thus, an appreciation of the potential for host-mediated immune damage and development of validated biomarkers that quantify the CSF immune response profile may inform judicious therapeutic selection and management to mitigate host damage in CNS infectious disease syndromes. Further dissection of associated inflammatory pathways may thus identify targeted interventions for prevention and/or treatment of these CNS infectious inflammatory syndromes.

ACKNOWLEDGMENTS

This research was supported by the Intramural Research Program of the National Institute of Allergy and Infectious Diseases (NIAID) at the National Institutes of Health (NIH).

The views herein do not reflect the official opinions of the Uniformed Services University or the Department of Defense.

We appreciate helpful discussions and manuscript review by I. Sereti and J. Bennett.

FUNDING INFORMATION

HHS | NIH | National Institute of Allergy and Infectious Diseases (NIAID) provided funding to Peter R. Williamson under grant numbers AI001124, AI001123, and AI109657. HHS | NIH | National Institute of Allergy and Infectious Diseases (NIAID) provided funding to Kim C. Williamson under grant numbers AI110852, AI069314, and AI114761. HHS | NIH | National Institute of Allergy and Infectious Diseases (NIAID) provided funding to David Boulware under grant number AI089244. HHS | NIH | National Institute of Allergy and Infectious Diseases (NIAID) provided funding to Anil A. Panackal under grant numbers AI001123, AI001124, and AI109657. HHS | NIH | National Institute of Neurological Disorders and Stroke (NINDS) provided funding to David Boulware under grant number NS086312.

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