Cryptococcal Meningitis

5

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Abbreviations

5-FC	Flucytosine, 5-fluorocytosine
ABLC	Amphotericin B lipid complex
AIDS	Acquired immune deficiency syndrome
AmB	Amphotericin B
ART	Antiretroviral therapy
CM	Cryptococcal meningitis
CMV	Cytomegalovirus
CNS	Central nervous system
CrAg	Cryptococcal antigen
CSF	Cerebrospinal fluid
CT	Computed tomography
CYP51	Cytochrome P51
ELISA	Enzyme-linked immunosorbent assay
GM-CSF	Granulocyte macrophage colony-stimulating factor
HIV	Human immunodeficiency virus
Hsp90	Heat shock protein 90
IDSA	Infectious Disease Society of America
IFN-γ	Interferon-γ
IL	Interleukin
IRIS	Immune reconstitution inflammatory syndrome
LA	Latex agglutination
LFA	Lateral flow assay
LFAmB	Lipid formulations of AmB

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MALDI-TOF-MS	Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry
MIC	Minimal inhibitory concentrations
MRI	Magnetic resonance imaging
PIIRS	Post-infectious inflammatory response syndrome
SOT	Solid organ transplantation
Th-1	T-helper type 1 response
TNF-α	Tumor necrosis factor-α
VP	Ventriculoperitoneal
WCC	White cell count
WHO	World Health Organization

Introduction

Ecology and Mycology of *Cryptococcus*

First identified in 1894, the genus of *Cryptococcus* comprises more than 30 known species, of which human infections are almost always caused by *Cryptococcus neoformans* and *Cryptococcus gattii* [1, 2]. Based on antigenic determinants on the polysaccharide capsule, the two varieties of *C. neoformans* are identified as *var. grubii* [serotype A] and *var. neoformans* [serotype D], while *C. gattii* includes serotypes B and C [3]. Recent genetic studies propose to redivide the two species into seven separate species and genotypes [4].

C. neoformans and *C. gattii* are encapsulated, heterobasidiomycetous fungi that exist in asexual or sexual stages [1]. *C. neoformans* was isolated from soil, avian excreta especially pigeons, and many other environmental sources, while *C. gattii* is restricted to red gum trees (*Eucalyptus*) [5–9]. The filaments that result from the mating of the two opposite types "*alpha*" and "*a*" have basidia that produce 1–2 micron basidiospores, thought to be the infectious propagules [10]. Most environmental and clinical isolates of *C. neoformans* only have the *alpha* mating locus shown to be more virulent in mice [11, 12]. This predominance can be explained by the yeast's ability under certain conditions to produce haploid fruiting without mating and sexual reproduction within the same mating type which may have explained the emergence of the Vancouver Island *C. gattii* outbreak [11–14].

Epidemiology and Risk Factors of Cryptococcal Meningitis (CM)

Cryptococcus is not considered a part of the human normal flora [15]. Prior to the era of acquired immune deficiency syndrome (AIDS), data analyzed from 725 isolates revealed that 100% of the isolates from Europe and Japan and more than 85% of the isolates from Canada, the UK, and the USA (except Southern California and Hawaii) were *C. neoformans* (serotypes A, D, or AD), while 35–100% of the isolates from tropical and subtropical areas were *C. gattii* (serotypes B and C). Overall,

C. neoformans serotypes were 86% of the isolates, and *C. gattii* serotypes were 13%, and 1% was not typeable [16].

Cryptococcosis remains a rare infection in normal hosts [15]. In fact, most adults and children in New York City were found to have antibodies to *C. neoformans* antigens, indicating that most of these infections are asymptomatic [17, 18]. In patients with AIDS, most infections are caused by *C. neoformans* serotype A [19], and *C. gattii* is much less common even in tropical and subtropical areas [20]. *C. gattii* is thought to cause disease predominantly in immunocompetent hosts, whereas *C. neoformans* mostly affects immunosuppressed patients [21], although *C. neoformans* (serotype A) in Vietnam has been associated with high prevalence of CM in human immunodeficiency virus (HIV)-negative, immunocompetent patients [22].

CM is the most common cause of adult meningitis in HIV patients in areas with high prevalence of HIV [23, 24]. The lower the $CD4^+$ count in HIV patients, the higher the incidence of cryptococcosis, and that skyrockets with $CD4^+$ count <100 cells/μL [25, 26]. The incidence of CM has declined significantly in Europe and the USA following the wide availability of antiretroviral therapy (ART) since 1997. Similarly, the rate of hospitalization in the USA declined from 16.6 million in 1997 to 7.7 million total population in 2009 [27, 28]. This was not seen in Africa as many patients present with a history of ART use and low $CD4^+$ count due to nonadherence and loss of follow-up [29]. The updated analysis of the global burden of HIV-associated CM in 2014 estimated the global annual rate of CM as 223,100 cases and global deaths of 181,100, of which 73% and 75%, respectively, were in sub-Saharan Africa [30]. This is, however, a remarkable reduction from 957,000 annual CM cases and 600,000 deaths estimated in 2009 [31].

In HIV-negative patients, most patients with disseminated cryptococcosis have an identifiable underlying disease. For example, these infections are seen in patients with hematologic malignancies, treatment with corticosteroids, sarcoidosis with or without corticosteroids, and solid organ transplantation (SOT) but not in bone marrow transplantation likely due to the routine use of azole antifungal prophylaxis in these patients. Other populations at risk are patients with abnormalities in cell-mediated immunity [32, 33].

Of note, 51% of HIV-negative patients with cryptococcosis had central nervous system (CNS) involvement, and of that 30% had no apparent predisposing conditions [34]. The "normal host" may actually have subtle or uncommon immune abnormalities [2]. Furthermore, smoking and outdoor occupations was associated with increased risk of cryptococcal infections in HIV-infected patients [35]. Table 5.1 shows common predisposing conditions for CM [34, 36–44].

Clinical Manifestations of Cryptococcosis

Pathogenesis, Immune Responses, and Neurotropism

After inhaling the aerosolized basidiospores from the environment, the immune system of a normal host can efficiently kill the yeast [1]. Alternatively, the initial possibly asymptomatic infection is contained in a primary complex in the hilar lymph

Table 5.1 Predisposing conditions to cryptococcal meningitis

Autoimmune disorders
• Sarcoidosis
• Systemic lupus erythematosus
Comorbidities
• Cirrhosis
• Diabetes mellitus
• Hepatic disease
• Lymphoproliferative diseases
• Peritoneal dialysis
Drugs
• Corticosteroids
• Monoclonal antibodies (adalimumab, alemtuzumab, infliximab)
Immunodeficiencies
• Chronic granulomatous disease
• FCg receptor II polymorphism
• GATA2 mutations
• Hyperimmunoglobulin E (Job syndrome)
• Hyper-IgM syndrome
Infections
• HIV infection
Syndromes and autoantibodies
• Autoantibodies to IFN-γ
• Autoantibodies to GM-CSF
• Idiopathic $CD4^+$ lymphopenia
• Pulmonary alveolar proteinosis
Transplantation
• Solid organ transplantation

nodes similar to primary tuberculosis [45]. This process involves $CD4^+$ T cells, interleukin-2 (IL-2), and tumor necrosis factor-α (TNF-α) [1, 46]. On the hand, the infection may disseminate outside the lungs in immunocompromised hosts and sometimes in normal hosts following a primary infection or a reactivation in dormant hosts after a decline of the $CD4^+$ count or the use of corticosteroids [1, 47].

C. neoformans has many virulence factors, of which the capsule is the most defined [48]. The polysaccharide capsule helps to evade phagocytosis by macrophages [49], activates the alternative complement pathway leading to depletion of complements [50], inhibits T-cell activation and pro-inflammatory cytokines such as TNF-α [51, 52], downregulates the antigen-presentation capacity of monocytes [53], and decreases the production of interferon-γ (IFN-γ) which suppresses the IL-12 production leading to inhibition of the protective T-helper type 1 response (Th-1) against *C. neoformans* [54, 55]. It also enhances HIV replication and resists oxidative stress [1, 48].

Other important virulence factors in *C. neoformans* which can explain the yeast neurotropism are (A) a laccase enzyme that converts CNS catecholamines to melanin that protects against oxidative stress and exerts multiple cell-wall functions [56], (B) thermotolerance of *C. neoformans* to high temperatures up to 43 °C compared to *C. gattii* and serotype D that do not tolerate heat above 40° [57], (C) a urease and metalloprotease Mpr1 enzymes in *C. neoformans* that facilitate its transcellular

migration into the mouse brain [58, 59], and (D) mechanisms in *C. neoformans* that allow it to survive nutrient starvation in the brain [60].

Pulmonary, Disseminated Disease and Atypical Sites of Infections

Cryptococcus causes a wide spectrum of infections with two major sites: the lungs and the CNS [15]. In immunocompetent hosts, pulmonary infections may be asymptomatic or may present with fever, chills, cough, chest pain, productive cough, hemoptysis, weight loss, and night sweats [61]. *C. neoformans* may colonize the respiratory tract of patients with chronic lung disease without underlying immune dysfunction. Infection may only involve the lungs associated with negative serum cryptococcal antigen (CrAg), but serum CrAg positivity should prompt ruling out an extrapulmonary focus of infection [62].

Most immunosuppressed patients present symptomatically, and pneumonia may progress faster and cause acute respiratory distress syndrome [63]. These patients may present with meningeal rather than pneumonia symptoms despite having both infections. Other coinfections have to be considered in AIDS patients with $CD4^+$ count <100 cells/μL especially cytomegalovirus (CMV), *Nocardia, Pneumocystis*, and typical and atypical mycobacteria [1, 64].

Cryptococcus can infect any organ system of the body. Noteworthy, skin involvement is almost exclusively associated with disseminated disease, and lesions can be of any type. Lesions may mimic bacterial cellulitis or abscess, acne vulgaris, molluscum contagiosum, and squamous or basal carcinoma and may originate deeper from the underlying bone or subcutaneous tissue [1, 65, 66]. Of note, SOT recipients on tacrolimus were found to have more skin and soft tissue infections than CNS infections. This may be explained by the antifungal activity of tacrolimus at 37–39 °C and the lower skin temperatures [67]. Another site of the infection is the prostate which is usually asymptomatic, and the isolation of *Cryptococcus* in the urine indicates disseminated disease [1]. Of note, the prostate may be a reservoir for the yeast which may grow in the urine even after the successful treatment of CM in AIDS patients [68].

CNS and Ocular Disease

CM may present with fever, headache, altered mental status, cranial nerve palsies, lethargy, coma, and memory loss [1, 15]. HIV patients with CM usually present after 2 weeks of the onset of symptoms and have a more disseminated disease, while non-HIV patients with CM may present after 6–12 weeks of the onset, a diagnosis often delayed by the absence of fever in non-HIV patients [2]. HIV patients with CM have more yeast burden, higher CSF CrAg titers, and higher rates of increased cerebrospinal fluid (CSF) pressure [1, 69]. In fact, 51% of HIV patients with CM have an opening pressure of >250 mm H_2O [70]. Also, HIV patients are also more likely to have other infections such as *Toxoplasma gondii* or CNS lymphomas [1]. Interestingly, C. *gattii* is associated with more cryptococcomas and hydrocephalus than *C. neoformans* [71].

Ocular disease is frequently seen in patients with CM. The most common findings are papilledema, cranial nerve palsies, and decreased visual acuity due to raised intracranial pressure [72, 73]. Visual loss may occur secondary to optic neuritis or endophthalmitis [74]. Furthermore, ocular coinfection may be seen with CMV and HIV [75]. In addition, compression of the ophthalmic artery may occur during the antifungal therapy due to raised intracranial pressure [1].

Outcomes and Prognostic Factors of CM

A study in Botswana showed no significant difference between the presentation and outcome in HIV-associated CM due to *C. neoformans* or *C. gattii* [76]. The updated analysis of the global burden of HIV-associated CM in 2014 estimated the 1-year mortality in patients in care to be 70% in low-income countries, 40% in middle-income countries, 20% in North America, and 30% in Europe, with 1.5 times higher mortality in patients not in care in these regions [30]. Risk factors that influence mortality in HIV-associated CM are CSF fungal burden, decreased sensorium, and the rate of clearance of infection [70]. In addition, HIV infection and cryptococcemia were associated with higher mortality rates, whereas hematologic malignancy and organ failure were not associated with mortality [77]. Also, low CSF white cell count (WCC) (<20 cell/μL) and high CSF CrAg titers >1:1024 were associated with worse outcomes [78].

In the USA, the mortality of HIV-negative patients was higher than HIV-positive patients (35% vs 26%) [77]. This may be attributed to the late presentation, delayed diagnosis, and possibly subtle immune dysfunction [77, 79]. Furthermore, the predictors of mortality of cryptococcosis in HIV-negative patients were shown to be age ≥60 years, hematologic neoplasm and organ dysfunction [34]. Also, a study of *C. gattii* CM in Australia showed that a CSF CrAg titer of ≥256 was associated with worse neurological consequences and death [80].

Cryptococcal Immune Reconstitution Inflammatory Syndrome (IRIS) in Patients with CM

Although the association between IRIS and CM in HIV patients is well established [81], IRIS has been described also in normal hosts, solid and bone marrow transplant recipients, and hematological malignancy patients on chemotherapy [1, 2, 82] after the immunosuppressive or antirejection regimens have been reduced to strengthen the immune system [83]. In apparently immunocompetent hosts, post-infectious inflammatory response syndrome (PIIRS) happens when cerebral edema and neurological damage are exacerbated by the immune response [79]. Two forms of IRIS identified in HIV patients are paradoxical IRIS in CM patients responding to antifungal therapy who relapse after initiating ART and unmasking IRIS in patients developing CM after starting ART [84].

IRIS may present with relapsing aseptic meningitis, abscess development, increased intracranial pressure, new focal findings, cryptococcomas, or other CNS

findings [85, 86]. Risk factors for CM-IRIS include high fungal burden which inhibits leukocyte migration into the CNS [87]; low initial CSF WCC and CSF protein levels as well as lower CSF IFN-γ, TNF-α, IL-2, IL-6, IL-8, and IL-17 cytokines; and higher CSF chemokines of macrocyte chemotactic protein-1, macrophage inflammatory protein-1α, and granulocyte macrophage colony-stimulating factor (GM-CSF). In addition, a rapid improvement of low $CD4^+$ cell count after starting ART is another major risk factor [88–91]. Predictors of IRIS in transplant patients include host immune responses and discontinuation of calcineurin inhibitors which causes a five times increased risk for IRIS [83]. The optimal time of starting ART and management options of IRIS will be discussed below in the management of CM section.

Diagnosis of CM

CSF Findings

HIV-negative patients with CM have increased CSF protein levels and WCC, while HIV patients have lower CSF protein levels and CSF WCC (median 15×10^6 cells/L) [70]. Low glucose levels and lymphocytic predominance are seen in both groups [2, 92]. India ink staining is a rapid tool for the diagnosis of CM and has a sensitivity of 50–70% in HIV-negative patients [93, 94] and a sensitivity/specificity of 84%/53% in HIV patients [95]. The performance of this test is highly operator-dependent.

CSF Cultures

Most bacterial and fungal media cultures of the CSF can detect the yeast in 3–7 days, with a sensitivity of 50–80% [93]. Biochemical reactions and DNA-based methods can help to identify isolates and distinguish between *C. neoformans* and *C. gattii* [96, 97]. Quantitative fungal cultures have been used to assess the rate of clearance and the fungicidal activity of various antifungal drugs [98]. Recently, matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS) has been studied and can rapidly identify species and genotypes of *Cryptococcus* [99].

Serum and CSF CrAg

Detection of cryptococcal capsular polysaccharide (glucuronoxylomannan, GXM) Ag in serum and CSF by latex agglutination (LA) and enzyme-linked immunosorbent assay (ELISA) has a sensitivity of 100% for disseminated disease and 94% for meningeal disease [100]. Specificity for CSF and serum CrAg was at least 90% for both LA and ELISA regardless of HIV status [101, 102]. In addition, cross-reactive serum CrAg has been seen in infections with *Trichosporon beigelii* [103] and *Stomatococcus mucilaginosus* [104].

A simple, quick, and cheap point-of-care test for the detection of CSF and serum CrAg has been developed; this new bedside lateral flow assay (LFA) has a sensitivity and specificity of 99% [105]. It has a preemptive role in resource-limited settings in the early diagnosis of asymptomatic CM and prevention of IRIS after starting ART [106] and in ART-naïve patients [107]. In addition, LFA's improved sensitivity offers an advantage over LA and cultures in diagnosing HIV-negative *C. gattii* meningitis [108].

Radiographic Findings

Brain magnetic resonance imaging (MRI) is more sensitive than computed tomography (CT) in CM, but there are no pathognomonic findings. Findings include lesions in the basal ganglia and midbrain that hyperenhance with T2-weighted images but do not enhance with T1-weighted postcontrast images [1]. Also, findings include hydrocephalus, single or multiple nodules with or without enhancement, dilated Virchow-Robin spaces, pseudocysts, masses, gyral enhancement, cryptococcomas, and lacunar and cortical infarcts [109, 110]. Even with the initiation of ART, these lesions may not resolve in months or years after successful treatment [111]; thus, cultures, symptoms, and clinical findings have to be considered before declaring treatment failure, and in the case of CNS parenchymal lesions, CNS lymphoma and coinfections with *Nocardia* or *Toxoplasma* should be ruled out [1]. Chest radiographs (chest X-ray, CT) can show single to multiple, well-defined noncalcified nodules in normal hosts diagnosed by lung biopsy. Other findings include lobar and mass-like infiltrates, hilar lymphadenopathy, lung cavities, and pleural effusions [112]. Disease may progress more rapidly in immunosuppressed patients such as AIDS patients or those receiving high-dose corticosteroids [63].

Management and Complications of CM

Antifungal Therapy

If untreated, CM can progress to altered sensorium, seizures, coma, and even death [2]. The rate of progression of the disease depends on host factors and fungal burden of *Cryptococcus*. The management of CM according to the practice guidelines of the Infectious Disease Society of America (IDSA) is based on three risk groups: HIV-infected individuals, organ transplant recipients, and non-HIV-infected nontransplant hosts [113]. The course is divided into three steps: induction, consolidation, and maintenance.

The use of Amphotericin B (AmB) has been imperative in the management of CM [114], and its combination with flucytosine (5-Fluorocytosine, 5-FC) was shown to be more fungicidal than AmB alone in sterilizing CSF [115]. The combination of AmB/5-FC was also associated with improved survival [116], reduced nephrotoxicity, shorter hospitalization [117], and prevention of relapse [118].

Patients with or predisposed to renal dysfunction or organ transplant recipients should not receive AmB deoxycholate (AmBd) but should be placed on lipid formulations of AmB (LFAmB) either with liposomal AmB (L-AmB) or AmB lipid complex (ABLC). Monitoring the kidney function is important during therapy with AmBd or LFAmB, and the dose of 5-FC has to be adjusted accordingly [113].

HIV-infected individual should receive induction with AmB/5-FC for at least 2 weeks until clinical response is seen, followed by consolidation with 400–800 mg fluconazole daily for 8–10 weeks and maintenance with 200 mg fluconazole daily for at least 1 year which can be stopped when CD4 count is ≥100 cells/μL, viral load is low or undetectable for ≥3 months, and serum CrAg is negative or low [113]. This requires the successful introduction of ART with the possibility of inducing paradoxical IRIS [113]. Current IDSA 2010 practice guidelines for CM recommend to start ART in 2–10 weeks after initiating induction, although more recent studies suggested 4–6 weeks as the most optimal time to start ART and prevent IRIS [70, 119]. Please see Table 5.2 for detailed recommendations of treatment of CM in HIV patients, SOT recipients, and HIV-negative patients.

5-FC is used in combination with one of AmB formulations for induction for at least 2 weeks (a dose of 100 mg/kg/day or renally adjusted) and should not be used alone as monotherapy can lead to resistance [120]. Monitoring of complete blood counts for bone marrow suppression is important, but it is not necessary to monitor serum drug levels [116, 121]. Also, monitoring of hypokalemia, hypomagnesemia, and acute kidney injury is essential in patients on AmB, and routine intravenous hydration and preemptive electrolyte replacement reduced the rates of hypokalemia and renal toxicity [122].

Azoles such as fluconazole have been used in the management of CM due to its safe profile and excellent penetration into the brain [123, 124]. However, due to its fungistatic properties, fluconazole should not be used in the induction phase when there is a high fungal burden in the CSF [1]. Itraconazole, although has less CSF penetration, was shown to successfully treat CM [125]. When 5-FC is not available, AmB plus fluconazole (800 mg/day superior to 400 mg/day) can be used [126]. Furthermore, fluconazole 1200 mg daily was shown to be more fungicidal than 800 mg daily in HIV-associated CM [127], and its combination with 5-FC (100 mg/kd/day) had early fungicidal activity close to that of AmB alone [128]. Also, voriconazole, posaconazole, and isavuconazole were used as salvage therapy in refractory cases with 38–60% response rates [129–131]. Echinocandins are not effective against *Cryptococcus* [132]. CNS cryptococcomas are treated similarly to CM but may require longer duration and surgical resection is rarely needed [133].

Persistence of CM Infection

Studies showed that in patients with AIDS and CM, at 10 weeks of therapy with AmB or fluconazole alone, 60–65% did not have a successful outcome compared to 35–45% failure rate in those who received a combination of AmB/5-FC or fluconazole/5-FC [134–137].

Table 5.2 Treatment recommendation of CM per IDSA 2010 guidelines

Stage	Regimen	Duration	Alternatives
Induction			
HIV patients	AmBd (0.7–1.0 mg/kg per day) plus 5-FC (100 mg/kg per day)	2 weeks	AmBd plus fluconazole, fluconazole plus 5-FC, fluconazole, itraconazole
	L-AmB (3–4 mg/kg per day) or ABLC (5 mg/kg per day) plus 5-FC (100 mg/kg per day)	2 weeks	–
	AmBd (0.7–1.0 mg/kg per day), L-AmB (3–4 mg/kg per day) or ABLC (5 mg/kg per day) alone in patient intolerant to 5-FC	4 weeks	–
Transplant recipients	L-AmB (3–4 mg/kg per day) or ABLC (5 mg/kg per day) plus 5-FC (100 mg/kg per day)	2 weeks	L-AmB (6 mg/kg per day), ABLC (5 mg/kg per day) or AmBd (0.7 mg/kg per day) all for 4–6 weeks
Non-HIV, nontransplant patients	AmBd (0.7–1.0 mg/kg per day) plus 5-FC (100 mg/kg per day)	≥4 weeks	–
	AmBd (0.7–1.0 mg/kg per day) alone in patient intolerant to 5-FC	≥6 weeks	–
	L-AmB (3–4 mg/kg per day) or ABLC (5 mg/kg per day) plus 5-FC (100 mg/kg per day) for AmBd-intolerant patients	≥4 weeks	–
Consolidation			
HIV patients	Fluconazole 400 mg daily	8 weeks	–
Transplant recipients	Fluconazole 400–800 mg daily	8 weeks	–
Non-HIV, nontransplant patients	Fluconazole 400–800 mg daily	8 weeks	–
Maintenance			
HIV patients	Fluconazole 200 mg daily	≥1 year	Itraconazole 400 mg daily (for ≥1 year)
			AmBd (1 mg/kg per week for ≥1 year)
Transplant recipients	Fluconazole 200–400 mg daily	6–12 months	–
Non-HIV, nontransplant patients	Fluconazole 200 mg daily	6–12 months	–

Persistence or relapse of infection may be difficult to identify but should be considered after at least 4 weeks of therapy with new signs or symptoms or repeat positive cultures and should not be based only on the persistence of positive India ink staining or CSF CrAg titers [1]. Also, a diagnosis of unmasking IRIS has to be considered in these settings. Most initial isolates of *C. neoformans* and *C. gattii*

have low minimal inhibitory concentrations (MICs) to AmB, 5-FC, and azoles by in vitro susceptibility testing [138]. Mechanisms of drug resistance in *C. neoformans* were described [139], and in fact, the clinical response may correlate with MIC levels [140]. Of note, molecular testing confirmed that most recurrent infections represented relapse of the initial strain rather than a new strain [141].

Management of IRIS in Patients with CM

The percentage of patients who develop IRIS (including paradoxical and unmasking) was shown to be 30% by 30 days of starting ART [142]. For patients who are worsening despite a sterile CSF, the IDSA guidelines recommend continuing antifungal therapy and using corticosteroids (0.5–1 mg/kg/day of prednisone equivalent) or dexamethasone at higher doses for severe CNS signs and symptoms [113]. Doses are to be tapered over the next 2–6 weeks. Nonsteroidal anti-inflammatory drugs and thalidomide were used but data is limited [113]. Also, recent cases suggest using TNF-α blockade with adalimumab in patients with severe CM-associated IRIS [143, 144]. Of note, chloroquine was used successfully to treat IRIS that resulted from withdrawal of corticosteroids due to its antifungal effects on *Cryptococcus* [145].

Management of Increased Intracranial Pressures in Patients with CM

Elevated intracranial pressure plays a critical role in the initial management of HIV-associated CM and improvement clinically and microbiologically [146]. Patients with severe CM often have CSF opening pressure >250 mm, acutely worsening brain edema, and possible development of CSF outflow obstruction [147]. Increased intracranial pressure may cause uncal herniation, tonsillar-cerebellar herniation, or compression of the midbrain [1]. During the early phase of treatment, controlling the increased intracranial pressure may be critical with external drainage by repeat lumbar punctures and ventricular or lumbar drains [148]. Hydrocephalus in patients with CM can be managed safely with placement of ventriculoperitoneal (VP) shunt even with positive CSF cultures [149]. Corticosteroids should only be used in patients with concomitant increased intracranial pressure and IRIS and not without IRIS [150].

Salvage and Adjunctive Immunomodulating Therapy

The use of adjunctive corticosteroids during initial combination antifungal treatment of CM was associated with increased disability, adverse side effects, and decreased rates of fungal clearance of CSF [150]. Stopping corticosteroids in SOT recipients is recommended to optimize immunity [83]; however, discontinuing calcineurin agents was associated with IRIS owing to the synergistic activity of

calcineurin inhibitors with antifungals against *Cryptococcus* [151]. The HIV-negative, apparently immunocompetent host may develop CM due to virulent strains; however, ruling our subtle immunodeficiencies (as outlined in Table 5.1) or idiopathic lymphopenia is recommended [152].

Adding sertraline to AmB and fluconazole has been associated with increased rates of CSF fungal clearance and less IRIS incidence and relapse [153]. Mycograb® which is a humanized Ab against heat shock protein 90 (hsp90) was shown to synergistically render AmB fungicidal and mirror the effects of 5-FC on the killing of *C. neoformans* [154]. Cytokine therapy with IFN-γ improved the fungal CSF clearance of *Cryptococcus* without increased adverse events [155]. Also, GM-CSF may enhance the anti-cryptococcal activity of monocytes and neutrophils in HIV patients [156, 157]. In addition, a new oral tetrazole, cytochrome P51 (CYP51) inhibitor Viamet 1129 showed potent in vitro activity against *C. neoformans* and *C. gattii* in animal models and may be used for fluconazole-resistant isolates [158, 159].

Screening and Prevention of CM

The goal of this approach is early diagnosis of HIV patients at high risk of developing CM through early detection of CrAg in the blood [106], detectable at median of 22 days before CNS symptoms [160]. The 100% negative predictive value supports its use in screening and preemptive fluconazole in CrAg-positive patients [2, 161]. In fact, the World Health Organization (WHO) supports its use in screening ART-naïve HIV patients with $CD4^+$ count <100 cells/μL and high prevalence of cryptococcal antigenemia (≥3%) [106, 162].

Research Gaps

Despite the use of combination therapy with AmB and 5-FC, CM mortality remains significantly high [30]. Many agents with potential antifungal properties remain under investigation [152]. A vaccine against cryptococcal GXM-tetanus toxoid conjugate was developed and elicited protective antibodies in mice [163]; however, human trials are yet to be conducted. The use of monoclonal antibodies could be promising but is to be further investigated and may require repeated injections [164]. Viamet 1129 is a new oral azole-like agent and may be promising but needs to be evaluated clinically. Further investigations of subtle immune deficiencies that predispose apparently immunocompetent hosts to cryptococcal infections may lead to the discovery of new agents and new mechanisms to better target and treat *Cryptococcus*.

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