Review

A neglected epidemic: fungal infections in HIV/AIDS

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Invasive fungal infections (IFIs) are a major cause of HIV-related mortality globally. Despite widespread rollout of combined antiretroviral therapy, there are still up to 1 million deaths annually from IFIs, accounting for 50% of all AIDS-related death. A historic failure to focus efforts on the IFIs that kill so many HIV patients has led to fundamental flaws in the management of advanced HIV infection. This review, based on the EMBO AIDS-Related Mycoses Workshop in Cape Town in July 2013, summarizes the current state of the-art in AIDS-related mycoses, and the key action points required to improve outcomes from these devastating infections.

Fungal infections and HIV/AIDS

Invasive fungal infections (IFIs) have rapidly emerged as a global threat to health owing to an increasing population of immunocompromised individuals and our dynamic interface with natural ecosystems [1]. The global HIV pandemic is now a major driver for mortality from fungal diseases worldwide [2]. Although combined antiretroviral therapy (cART) has dramatically changed the face of the HIV epidemic, many patients still present to clinical services with advanced HIV-related immunosuppression, particularly in developing countries. A failure to adequately address the IFIs that affect such patients has become a primary driver for AIDS-related death worldwide (Table 1). In July 2013, leading researchers from around the world working at the interface of HIV and IFIs met at the EMBO AIDS-Related Mycoses Conference in Cape Town, South Africa, to address these issues. In this review we summarise the major topics discussed: the epidemiological interaction between HIV and fungal infections; the immunopathogenesis and clinical aspects of fungal infection in the context of HIV/AIDS; and key knowledge gaps that need to be addressed to reduce the unacceptably high mortality. A formal position statement produced at the conference also accompanies this article [3].

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Common fungal infections in HIV and their global impact

Since the first cases of AIDS were identified in San Francisco and New York in the early 1980s, opportunistic fungal infections have been a primary driver for mortality from HIV infection. Although *Pneumocystis* pneumonia (PCP) was initially responsible for over 70% of the first 400 recorded deaths from HIV/AIDS, cryptococcal meningitis (CM) now accounts for the majority of worldwide deaths from HIV-related fungal infection (Table 1) [4,5]. Furthermore, the ongoing HIV pandemic has led to the emergence of further opportunistic infections in the context of endemic mycoses.

Cryptococcosis

CM is a devastating infection associated with a high case fatality rate, primarily associated with advanced HIV disease (typically a CD4 T cell count < 100 cells/mm^3) [5,6] and caused by the basidiomycete *Cryptococcus neoformans*. Infection is acquired by inhalation, and a failure to control latent infection in alveolar macrophages as a consequence of HIV infection leads to systemic dissemination with death from meningoencephalitis and raised intracranial pressure [5]. The majority of HIV-related CM occurs in sub-Saharan Africa; an epidemiological study published in 2009 suggested that there are 720 000 cases per annum in this region, and approximately 950 000 cases globally per annum [5]. There are an estimated 120 000 cases per annum in South and Southeast Asia, 7800 cases in North America, 6500 cases in North Africa and the Middle East, 500 cases in Western and Central Europe, and 100 cases in Oceania. As a consequence of these infections, there are 625 000 deaths due to CM globally per annum. Case fatality rates among treated patients vary widely, from approximately 9% in developed countries to 70% in sub-Saharan Africa, reflecting differences in time to diagnosis and therapy used. In addition, cryptococcal immune reconstitution inflammatory syndrome (IRIS) has emerged as a major problem and a significant contributor to mortality. We urgently need better global surveillance to accurately define the evolving disease burden of CM.

Amphotericin B and flucytosine are the cornerstone of antifungal therapy for CM. Recent studies have sought to determine optimal treatment regimens that can be used in developing countries, where amphotericin B is difficult to administer owing to its intravenous formulation, toxicity,

Table 1. Global burden of disease in the HIV–mycoses epidemic[a]

Invasive fungal disease	Main epicentres	Estimated cases per annum	Estimated mortality per annum	Refs
Cryptococcal meningitis	Sub-Saharan Africa, Southeast Asia	950 000	625 000	[2,3]
Pneumocystis pneumonia	Asia, Latin America, sub-Saharan Africa	400 000	150 000	[2,6]
Disseminated histoplasmosis	North America, sub-Saharan Africa	300 000	10 000	[17,19]
Disseminated penicilliosis	Southeast Asia	50 000	5000	[21]

[a]The data presented are based on the best available estimates, with relevant references as indicated. Notably, the data are likely to underestimate the true burden of disease, because they only represent estimates for the areas indicated, with a substantial number of sporadic cases occurring outside of the epidemic zones.

and lack of monitoring capacity, and flucytosine is not widely available. A large Vietnamese randomised controlled trial recently demonstrated a 39% reduction in 10-week mortality with amphotericin B and flucytosine versus amphotericin B alone for CM [7]. Further studies indicate that short courses of amphotericin B plus high-dose fluconazole, or triple combination therapy with short courses of amphotericin B, flucytosine, and high-dose fluconazole, are safe and rapidly sterilising [8]. Further clinical trials to determine regimens with the highest early fungicidal activity are ongoing.

Pneumocystis pneumonia

Pneumocystis jirovecii is an ascomycete yeast that came to prominence as the major pulmonary infection associated with advanced HIV disease during the early 1980s. Infection is characterised by progressive alveolitis leading to respiratory failure. Diagnosis of PCP requires radiographic and microbiological confirmation of disease; thus, the infection is generally treated empirically in resource-poor settings. This has led to underestimation of the true incidence of PCP in sub-Saharan Africa, and underscores the urgent requirement for point-of-care diagnostics to enable early identification of cases and accurate definition of the global disease burden [9].

A global meta-analysis of PCP demonstrated a positive correlation between gross domestic product and likelihood of PCP, consistent with a relatively lower incidence of PCP in some African countries, or possibly reflecting ascertainment bias in resource-poor settings [9]. Furthermore, PCP continues to be a major problem in children with perinatally acquired HIV worldwide [10]. Climatic factors are significant predictors of the likelihood of admission with HIV-related PCP [11], and PCP can be detected in the air near PCP-infected individuals. There have been several documented PCP outbreaks, further suggesting person-to-person transmission [12].

High-dose cotrimoxazole (trimethoprim–sulfamethoxazole combination therapy) is cheap, oral, and an effective first-line therapy for PCP. Optimal second-line therapy, often required owing to cotrimoxazole intolerance, should be clindamycin plus primaquine [13]. Cotrimoxazole prophylaxis in HIV patients with CD4 T cell counts of < 200 cells/mm^3 has significantly reduced the incidence of PCP and toxoplasmosis; it is safe to discontinue primary prophylaxis for PCP when the patient viral load is suppressed and the CD4 count is > 100 cells/mm^3 [14].

Clinical and laboratory factors (patient age, hypoxaemia, C-reactive protein, injection drug use, presence of pulmonary Kaposi's sarcoma, medical comorbidities, and PCP recurrence) are associated with worse outcomes in PCP [15]. These risk factors have been used to develop prognostic scoring tools that enable accurate identification of patients at higher risk of death, who require intensive inpatient management [16]. Adoption of such tools, in combination with point-of-care diagnostics, in resource-poor settings has the potential to enhance safe management of patients with PCP.

Oropharyngeal and oesophageal candidiasis

HIV is primarily associated with oropharyngeal candidiasis [17]. Three forms of mucosal candidiasis are common: pseudomembranous, erythematous, and oesophageal candidiasis. Infection prevalence increases greatly with CD4 counts < 200 cells/mm^3 [17]. A recent US study suggests a prevalence of 27% for oropharyngeal candidiasis in newly diagnosed HIV patients [17]. It has been shown that oropharyngeal candidiasis has an overall prevalence of approximately 10% in HIV-1 infected individuals in Asia, Africa, and Latin America [18]. cART significantly reduces the risk of oral candidiasis in HIV-1 infection; however, smoking appears to increase the risk [17]. Judicious use of topical nystatin or azoles (mainly fluconazole) in combination with cART has been the mainstay of therapy [17], although azole resistance may occur.

Endemic and other fungi

Disseminated histoplasmosis was first recognised as an AIDS-defining infection in 1987, and is present in up to a quarter of patients with advanced HIV-1 in endemic regions such as Indiana, USA. This led to the development of antigen detection assays for urine and blood, with sensitivity of the urine antigen test greater than 95% in HIV-positive individuals. The vast majority of cases of histoplasmosis occur when HIV-positive individuals have CD4 T cell counts of < 100 cells/mm^3 [19]. Fortunately, response rates to therapy tend to be good, with an 85% success rate for itraconazole and 98% for amphotericin B [20]. Secondary prophylaxis is required until CD4 T cell counts are > 150 cells mm^3.

Climactic variables such as rainfall are predictive of disseminated histoplasmosis, suggesting that some cases are due to recent exposure rather than reactivation of latent disease [21]. *Histoplasma duboisi* has increasingly been recognised as a disseminated opportunistic mycosis in the context of advanced HIV infection in Africa. Histoplasmosis is estimated to account for approximately 2% of AIDS presentations in sub-Saharan Africa [22]. A recent study in Cape Town, South Africa, further demonstrated that a significant proportion of histoplasmosis-like infections are due to *Emmonsia* spp. [23].

Penicilliosis is an endemic infection in Southeast Asia that is now the third most common AIDS-defining illness in that region [24]. Bamboo rats are an important natural

host, and human disease occurs primarily during the rainy season. Most patients with disseminated penicilliosis present with CD4 counts of < 50 cells/mm^3 [25].Common features include fever, skin lesions, hepatomegaly, lymphadenopathy, weight loss, and cough. Overall mortality is approximately 10% in hospital settings, with 75% cure rates with either itraconazole or amphotericin B [24].

Pulmonary aspergillosis has been a historically common AIDS-related infection, and occurred mainly secondary to AZT-related neutropenia or corticosteroid use. However, sporadic cases may occur in the context of profound HIV-related immunodeficiency [26]. Chronic pulmonary aspergillosis may now be emerging as a complication of lung damage in HIV–tuberculosis. Given the difficulties in the diagnosis of pulmonary aspergillosis, development of simple diagnostic tools will be critical to defining the scale of this problem.

HIV/AIDS: consequences for antifungal immunity

Although a decline in peripheral blood CD4 count has historically been a robust predictor of risk for opportunistic fungal infection in HIV [27], there is now increasing recognition that complex effects of HIV infection in both myeloid and lymphocytoid lineages contribute to increased susceptibility to IFIs (Figure 1).

T cell dysfunction in HIV

HIV-1 infection leads to rapid destruction of memory CD4 T cell clones, and ultimately the failure of infection-specific effector populations [27]. It has been shown that early HIV infection significantly depletes gut T helper 17 (Th17) cells, leading to susceptibility to mucosal candidiasis [28], and mirrors immune deficits observed in chronic mucocutaneous candidiasis. Alveolar T cells from HIV-1 infected individuals also exhibit impaired antigen-specific cytokine responses for respiratory pathogens [29]. Thus, HIV appears to facilitate mucosal candidiasis through Th17 cell depletion.

In patients receiving cART, mucosal Th17 numbers are rapidly restored; however, there is a specific failure to normalise central and memory CD4 T cells [30]. STAT4-dependent protective Th1 and Th2 responses in the lung are also important for immunity to PCP [31], and HIV-infected individuals colonised with PCP have lower Th2 cytokines than control subjects [31]. Impaired CD4 T cell responses to cryptococcal mannoprotein are strongly

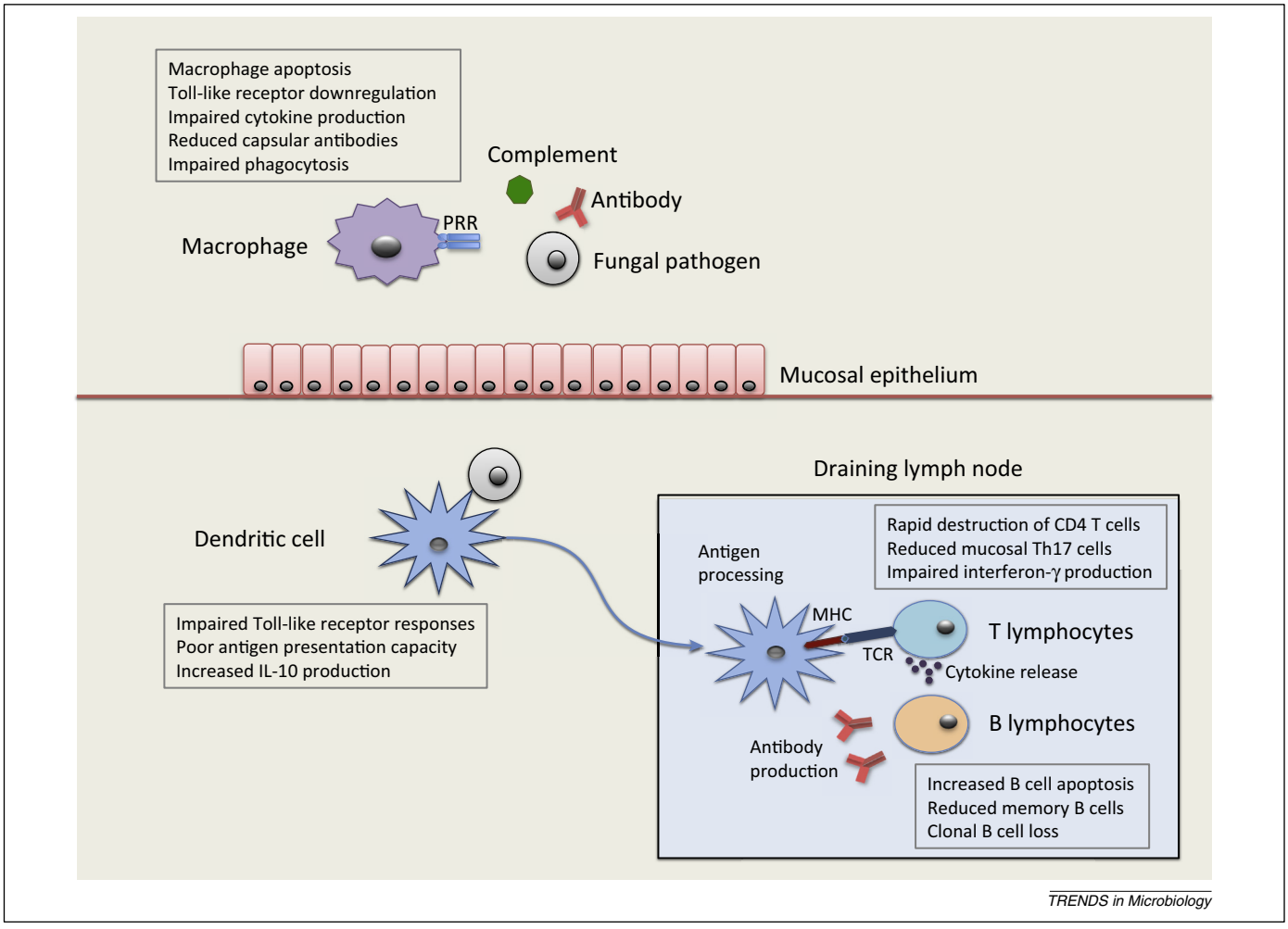

Figure 1. Impact of HIV infection on cellular immunity to opportunistic fungal pathogens. HIV causes multiple defects to both innate and adaptive arms of the immune system, leading to heightened susceptibility to both mucosal and invasive fungal diseases. Infection of macrophages by HIV leads to impaired fungal processing and cytokine production. Dendritic cell responses are dysregulated, with poor antigen presentation to T lymphocytes. T lymphocyte numbers are reduced, with specific defects in mucosal Th17 responses. Loss of clonal B cell memory subsets further contributes to impaired responses, especially to capsulated organisms. More detailed descriptions of the precise defects in fungal immunity are available elsewhere [24,28,29,31,39,46]. Abbreviations: PRR, pattern recognition receptor; TCR, T cell receptor; MHC, major histocompatability complex.

associated with a failure to produce interferon-γ or tumour necrosis factor-α (TNF-α) and poor CM outcome. Furthermore, competent responses are associated with reduced cryptococcal burden in cerebrospinal fluid (CSF), higher CSF lymphocyte counts, and a faster rate of sterilisation [6]. Clinical studies have revealed a complex relationship between cryptococcal genotype, mortality, capsule shedding, and Th1/Th2 host responses [32]. Therefore, it appears that competent polyfunctional T cell responses are crucial for optimal sterilising antifungal immunity in HIV.

The antibody response in HIV

Infection with HIV-1 leads to impaired B cell function, with a reduction in the proportion of resting memory B cells and emergence of abnormal B cell populations in blood [33]. These changes correlate with the degree of viraemia and the CD4 T cell nadir. HIV-dependent T cell activation in the lymph nodes also leads to impaired B cell development in germinal centres [34] and an increase in an activated subset of memory B cells that are sensitive to Fas-mediated apoptosis [35]. In viraemic or lymphopoenic HIV-1-infected individuals, Fas expression is extensively increased on all B cell subsets, and appeared to be associated with T cell activation [35]. Notably, there is limited impact of cART on resting memory B cell depletion [33].

The past decade has witnessed a major resurgence in interest in antibody-mediated protection against fungal infection [36]. Studies in CM suggest that capsular antibodies may be able to inhibit yeast budding [37], although major variation in fungal serotype specificity could complicate further translation. Murine studies further demonstrate a clear role for B cell B-1 B subpopulations in the early innate control of *C. neoformans* infection [38]. In addition, a critical role has been demonstrated for serum IgM in the control of cerebral cryptococcosis [39]. A clinical study demonstrated that PCP-infected patients developed IgG responses to KEX1 and major surface glycoproteins. Furthermore, low KEX1 IgG levels were independently predictive of PCP risk [40]. A class of natural IgM antibodies have been identified in mice and are specific for the fungal cell wall components chitin and β-glucan [41]. These antibodies mediate recruitment of dendritic cells (DCs) to the lung in murine PCP and drive protective adaptive responses. Furthermore, natural IgM antibodies with similar specificities are present in fish and primates, suggesting an evolutionarily conserved role in fungal immunity [41]. These observations support the concept of common fungal antibody targets that could be exploited for immunotherapy [36].

Myeloid function in HIV

Myeloid cells are a major reservoirs of HIV infection [42]. However, the effects of HIV-1 infection on innate antifungal immunity have been poorly defined. HIV-1 infects macrophages at a relative low frequency, leading to expansion of 'non-classical' monocytes, downregulation of Toll-like receptor 4 (TLR4) and CD14, impaired NF-κB-dependent cytokine production [43], increased production of IL-10 [43], and TRAIL (TNF-related apoptosis-inducing ligand) decoy receptor-mediated apoptosis through intracellular c-FLIP (cellular FLICE-like inhibitory protein) [44]. Furthermore, phagocytosis of *Aspergillus fumigatus* and *P. jirovecii* is impaired and mannose receptor expression is reduced [43]. Zymosan stimulation leads to inhibition of HIV-1 replication in macrophages through IL-10-dependent mechanisms [45]. Conversely, exposure of HIV-1-infected DCs to *Candida albicans* leads to increased HIV-1 replication and higher levels of IL-10/IL-1β [46].

It has also been shown that HIV-1 has direct and indirect effects on DC function, leading to dysregulation of both myeloid and plasmacytoid subsets [47]. This results in impaired TLR responses and poor antigen presentation capacity [47]. Furthermore, infection of DCs with *Penicillium marneffei* promotes endocytosis of HIV-1, leading to enhanced trans-infection of CD4 T cells by HIV-1 [48]. Clinical studies in HIV cohorts indicate an association between polymorphisms in the DC trafficking chemokine receptor CCRL2-167F and rapid progression to PCP, consistent with a role for DC recruitment to the lung for control of PCP infection [49].

Clinical studies have also demonstrated that capsule size is a driver for raised intracranial pressure in CM. Interestingly, increased capsule size was associated with reduced inflammatory responses in CSF, suggesting an immunosuppressive effect for cryptococcal capsules within the macrophage phagolysosome [32]. Taken together, all these observations suggest that HIV infection leads to quantitative and qualitative defects in innate antifungal immunity, and that fungal co-infection modulates the host immune responses to facilitate heightened HIV replication.

Treatment options and the way forward

Despite major progress in both translational research and drug development in the field of medical mycology in recent years, both the incidence and mortality of invasive fungal infections in HIV/AIDS remain unacceptably high in developing countries. Better access to existing therapies, new therapies with better tolerability and efficacy, point-of-care diagnostics, enhanced surveillance, improved management of immune dysfunction, and effective fungal vaccines are all required to improve outcomes.

New drugs

There are major pragmatic barriers for the development of effective antifungal therapy: because fungi are eukaryotic pathogens, drug development is limited by a lack of fungal-specific drug targets; the existing lipophilic triazoles, polyenes, and echinocandins have poor or no oral bioavailability; clinical failure is high owing to the immunocompromised status of the host and the development of resistance for certain drugs; and toxicity can occur owing to off-target effects on the host. The development of novel drug classes is urgently required to improve the unacceptably high mortality (Table 2).

Although cheap and effective treatment options such as oral fluconazole and itraconazole are readily available for diseases such as mucosal candidiasis and endemic mycoses, better oral drugs with satisfactory tolerability are required, in particular for CM and to a lesser degree for PCP. At the EMBO AIDS-Related Mycoses Workshop,

Table 2. A medical mycology toolkit to fight the global AIDS–mycoses epidemic

Tool required	Diseases targeted	Global impact
Point-of-care diagnostics	*Pneumocystis* pneumonia, disseminated histoplasmosis, disseminated penicilliosis	Enhanced diagnosis, early therapy, reduced mortality, better surveillance
Disposable spinal needles and manometers	Cryptococcal meningitis	Improved patient management, reduced mortality
New orally active agents	Cryptococcal meningitis	Improved patient survival, earlier patient discharge, reduced drug toxicity
Vaccines and immunotherapy	Cryptococcal meningitis, *Pneumocystis* pneumonia, histoplasmosis, penicilliosis	Reduced latent infection, enhanced antifungal immunity, disease prevention, improved outcomes from infection
Improved access to existing medicines	Cryptococcal meningitis	Improved outcomes from infections, reduced drug toxicity
Global surveillance	Cryptococcal meningitis, *Pneumocystis* pneumonia, histoplasmosis, penicilliosis	Increased understanding of global burden, better targeting of resources, increased insight into risk factors

access to flucytosine for CM was also identified as a major issue in developing countries [50]. Given the difficulties with administration of intravenous amphotericin B, fluconazole is widely used for induction therapy in sub-Saharan African countries, leading to increased risk of death from CM [50]. Unfortunately, since the development of the echinocandins and second-generation triazoles (all prohibitively expensive in developing countries) there has been a major shortfall in the development of new antifungal drug classes.

Another critical issue is around the management of raised intracranial pressure in CM, which is a key cause of mortality. Large-volume CSF drainage is often repeatedly required to normalise intracranial pressure; however, this requires disposable spinal needles and manometers, which are not widely available [32]. A key goal would be to enable global access to these simple medical devices through well-functioning supply chains (Table 2).

Given the abundance of β-(1,3)-glucan in the cell wall of *Pneumocystis*, clinical studies have addressed the potential utility of echinocandin β-(1,3)-glucan inhibitors in this infection [51]. However, there are currently no good randomised controlled trial data to support their use. Initial studies of echinocandin efficacy in mice and rats with PCP indicate improved survival with echinocandin treatment, and it was subsequently shown that this is due to specific activity against the cyst form of PCP [52]. Murine studies further support the efficacy of low-dose caspofungin when combined with cotrimoxazole for the treatment of PCP [53]. However, echinocandins are currently very expensive and only available as intravenous formulations, and thus are not suitable for use in many developing countries. A further target would be to improve access to safer formulations of amphotericin B and flucytosine.

Rapid diagnostic tests for fungal infection

Initial studies in South Africa showed that cryptococcal antigen screening in asymptomatic individuals with HIV followed by high-dose fluconazole treatment is a cost-effective strategy for reducing mortality associated with CM [54]. A new point-of-care dipstick assay has high positive correlation with validated antigen assays, and allows screening to be performed on plasma, serum, urine, or venous blood in resource-poor settings [55].

There is a strong correlation between blood β-glucan levels and HIV-related PCP [56]. However, the β-glucan assay is not specific for PCP, and is expensive and difficult to use in resource-poor settings. Quantitative PCR for PCP is highly sensitive and specific for nasopharyngeal aspirates, raising the possibility that minimally invasive airway sampling could be a viable diagnostic route in resource-poor settings [57]. However, a cheap point-of-care diagnostic that is easy to use is urgently required for PCP.

Rapid diagnosis of histoplasmosis represents a further logistical problem, because prolonged culture may be required and the current gold-standard serological assay (MiraVista Diagnostics, Indianapolis, USA) is not commercially available. Commercial urine-based antigen tests have recently been developed, approved by the FDA, and evaluated. Initial studies indicate reasonable sensitivity and specificity [58]. Likewise, diagnosis of penicilliosis is primarily reliant on culture-based methods, although there has been some recent progress with serodiagnostic and PCR-based methods [59].

Impact of highly active antiretroviral therapy (HAART) roll-out

cART roll-out in the developed world over the past 15 years has clearly led to major reductions in the incidence of HIV-related opportunistic infections such as oropharyngeal candidiasis, PCP, and CM [60]. It has also been shown that cART roll-out is associated with reduced incidence of CM in African settings [61]. These observations provide a compelling argument for the acceleration of current programmes to improve access to cART for all. However, given the issues around increased mortality from cryptococcal IRIS and early cART, careful screening and identification of patients with CM will be required.

Cryptococcal IRIS

Cryptococcal IRIS has emerged as a major cause of morbidity and increased risk of death in patients with HIV-associated CM [62]. Cohort studies indicate that IRIS is associated with high levels of serum cryptococcal antigen and pre-IRIS non-Th-1 cytokine profiles in peripheral blood [62]. Patients with cryptococcal IRIS have increased proportions of CCL2/CXCL10 and CCL3/CXCL10 on CSF CD4/CD8 T cells and higher ratios of $CXCR3^{+}CCR5^{+}CD8^{+}$ T cells compared to patients not developing IRIS, suggesting that

$CD8^+$ T cells and myeloid cell trafficking may be involved in this syndrome [63]. Early cART (within 7 days) is associated with increased risk of cryptococcal IRIS when compared to delayed cART (>28 days) [64]. A systematic review suggested that the optimal timing for cART in CM is unclear, mainly owing to a lack of mortality-based evidence; however, it is recommended that cART be delayed until there is clinical evidence of response to antifungal therapy [65]. Current studies of steroid usage in CM may yield a better understanding of their prophylactic utility for CM IRIS.

It has been historically shown that steroids are a highly beneficial adjunctive therapy for pulmonary inflammation and hypoxaemia associated with severe PCP. This suggests that the host response plays a significant role in the pathogenesis of alveolitis. A recently developed dectin-1 carbohydrate-binding domain fusion to murine FC antibody fragment regions has high affinity for PCP β-glucan and reduced fungal burden in animal models [66]. In a murine model of PCP IRIS, T cell recovery was associated with severe pulmonary inflammation and increased phagocytosis by alveolar macrophages [67]. Treatment with the immunomodulatory drug sulfasalazine lead to attenuation of IRIS associated with Th2 polarisation in the lung and alternate activation of macrophages [67]. Depletion of T cells in mice with PCP IRIS using anti-CD3 antibodies led to striking improvements in survival and reductions in BAL CD4/CD8 cells [68]. However, HIV-related PCP IRIS is rare clinically.

Cutaneous IRIS has also been reported as a consequence of *P. marneffei* infection in Thai HIV-1 patients on cART initiation [69]. *Histoplasma* IRIS has also been reported, associated with a range of manifestations including the haemophagocytic syndrome [70].

Immunotherapy and vaccines

There has been considerable interest in augmenting host Th1 responses to fungal infection. A recent randomised controlled study of adjunctive recombinant interferon-γ therapy in CM in combination with amphotericin B and flucytosine demonstrated increased fungicidal activity in CSF and good tolerability; however, there was no significant difference in mortality in this initial study [71]. Studies are now ongoing to evaluate the utility of adjunctive steroid therapy in CM (http://www.controlled-trials.com/ISRCTN59144167/).

Further studies are evaluating the potential of the 18B7 monoclonal antibody for radioimmunotherapy of CM [72]. A H99 *C. neoformans* strain expressing interferon-γ has been developed and assessed for its potential as a vaccine [73]. In BALBc murine infection models, immunisation with this strain led to enhancement of pulmonary sterilising responses, associated with enhanced granuloma formation and faster resolution of inflammation in the lung [73]. Further studies indicate protection was dependent upon competent Th1-mediated immune responses [73].

However, there are still no vaccines for any fungal pathogen that have been developed for clinical use. However, there are considerable challenges to developing efficacious vaccines that work in severely immunocompromised hosts, for whom direct cellular or antibody-mediated therapy is likely to be required. Further studies in HIV patients with insufficient immunological reconstitution after cART have demonstrated the potential of umbilical cord mesenchymal stem cell therapy to augment competent CD4 T cell responses [74]. However, deployment of such techniques in resource-poor settings is not currently feasible.

Box 1. Outstanding questions

- What is the true global burden of fungal disease in HIV?
- How is cART roll-out impacting opportunistic fungal infections?
- How many lives can be saved through roll-out of cheap diagnostics and better drug access?
- How useful will immunotherapy and vaccines be for opportunistic fungal infections in HIV patients?
- What is the best strategy for treatment and management of cryptococcal IRIS?
- Is myeloid dysfunction a major driver for susceptibility to opportunistic fungal infection in HIV?

Concluding remarks

IFIs have emerged as a serious driver of worldwide mortality in the context of the global HIV pandemic. Despite a major scientific effort to find new cures and vaccines for HIV, hundreds of thousands of HIV-infected individuals continue to die on a yearly basis from secondary fungal infection. We urgently need to better understand the underlying immunopathogenesis of fungal infection in HIV to enable better diagnosis and management in at-risk populations (Box 1). There is also a pressing and pragmatic requirement for access to effective drugs and other therapies in developing countries. In addition, the development of novel diagnostics to enable early diagnosis of fungal infection is required for improved outcomes and better epidemiological definition of the fungal disease burden worldwide.

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