Curr Fungal Infect Rep (2016) 10:153–162
DOI 10.1007/s12281-016-0267-0

FUNGAL GENOMICS AND PATHOGENESIS (S SHOHAM, SECTION EDITOR)

Pathogen–Host Interaction of *Histoplasma capsulatum*: an Update

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Published online: 21 November 2016

Abstract
Purpose of Review Histoplasma capsulatum is a dimorphic fungus endemic to the Americas but is increasingly recognized as a global pathogen. In this review, we describe the most current findings in host evasion, host–pathogen interaction, therapeutics, and vaccines.
Recent Findings Recent advances in the understanding of *H. capsulatum* virulence and immunity include the importance of host–pathogen nutrient modulation, immune cell transcriptional regulators, cytokine signaling, and coordination of innate and adaptive immunity.
Summary The latest developments in our understanding of *H. capsulatum* infection lay the foundation for future clinical breakthroughs in prevention, diagnosis, and treatment of this intracellular fungal pathogen.

Keywords *Histoplasma capsulatum* · Fungal disease · Pathogen–host interaction · Innate immunity

This article is part of the Topical Collection on *Fungal Genomics and Pathogenesis*

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Introduction

Histoplasma capsulatum is the most common endemic pulmonary mycosis infection in the USA [1]. Although found worldwide, *H. capsulatum* is endemic to the Ohio River and Mississippi River valleys in the USA. It is a thermally dimorphic fungus that grows in hyphal form in soil and once inhaled into lungs, converts to the pathogenic yeast form. Infection can range from asymptomatic or mild flu-like symptoms to deadly disseminated disease. Up to 25,000 people develop life-threatening histoplasmosis in the US endemic regions every year [2]. Host clearance requires coordination between innate and adaptive immune responses. This review will describe the pathogenicity of *H. capsulatum*, immune response to the fungus, and potential new therapies to combat infection, focusing on pathogen–host interaction in the past 3 years of literature.

Pathogenesis From the Perspective of *H. capsulatum*

H. capsulatum is a primary fungal pathogen. The course and outcome of disease are determined by the interaction between the fungus and host cells. It exhibits a number of attributes that enhance its survival upon contact with phagocytes (Fig. 1a).

H. capsulatum Invasion

H. capsulatum utilizes multiple strategies to avoid attack by immune cells. These can be categorized as masking detection and mitigating attack. Eight clades of *H. capsulatum* isolated geographically have been classified into chemotype I or II based on the absence (chemotype I) or presence (chemotype II) of α-(1,3)-glucan. The most well-studied clinical strains of *H. capsulatum* G217B and G186A belong to chemotypes I

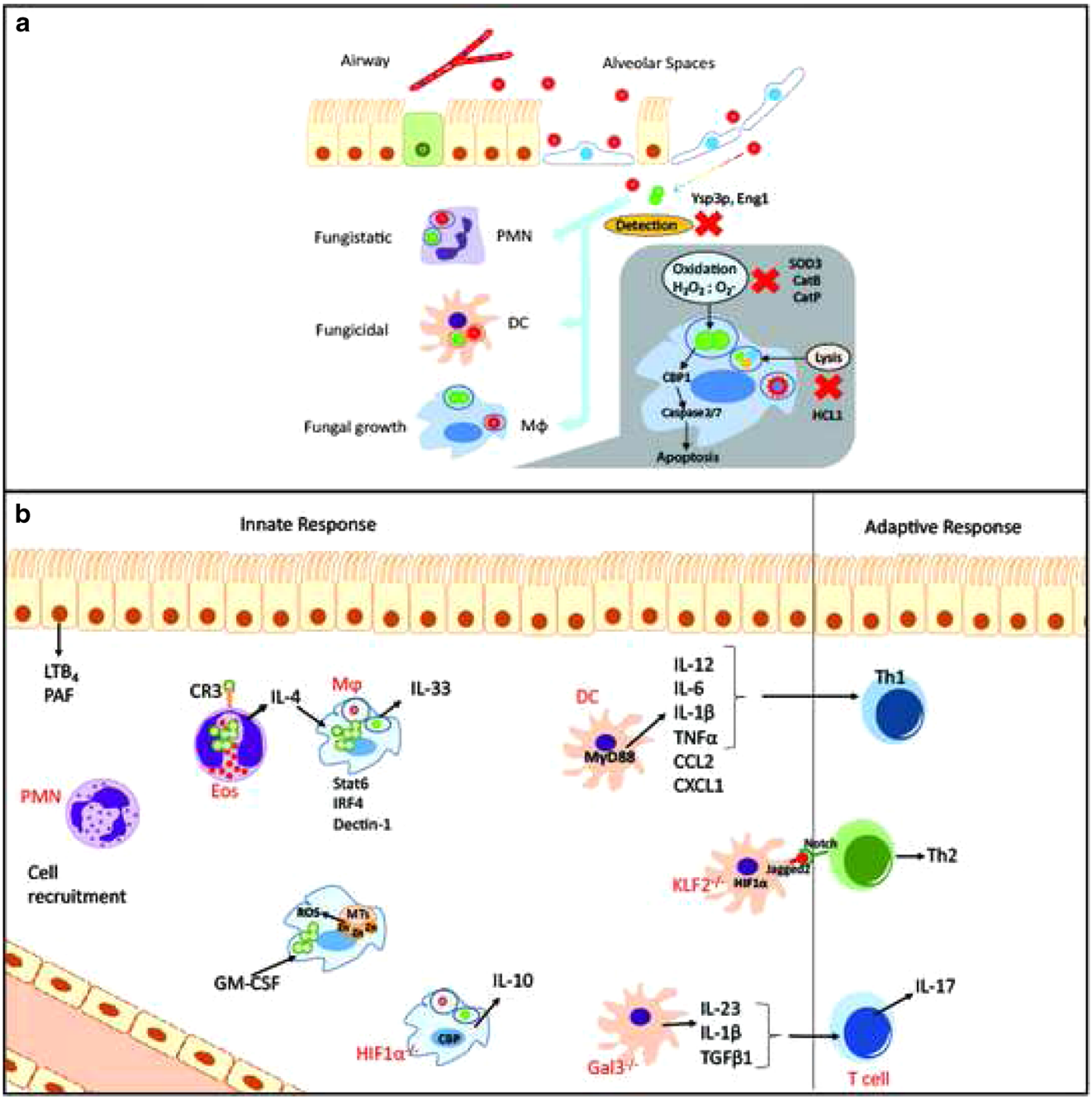

Fig. 1 **a** The illustration of pathogenesis of *H. capsulatum*. The mycelia fragments and conidia travel down airway to terminal alveolar where the conidia pass through the loose structure of epithelium. The temperature of hosts converts conidia to yeast. To avoid detection, yeasts produce proteins such as Ysp3p, Eng1 to mask the exposure of β-glucan. Phagocytes that have taken up conidia/yeasts either inhibit yeast growth (PMN), kill the yeasts (DC), or allow yeast growth (Mφ). Yeasts in Mφ can stimulate the production of proteins (e.g., SOD3, CatB, CatP, etc.) to reduce oxidative damage and prevent lysis. **b** The immune response to *H. capsulatum*. To combat *H. capsulatum* infection, innate cells are recruited to the site of infection by chemoattractants secreted by epithelial cells and other immune cells. After migrating to the lungs, Mφ need to be activated by proinflammatory cytokines to halt fungal growth. Inappropriate Mφ stimulation results in uncontrolled *H. capsulatum* expansion. The proper T cell activation relies on DC activation to initiate adaptive immunity, coordinating the innate and adaptive responses to clear the fungus

and II, respectively. Production of α-(1,3)-glucan is a strategy of chemotype II to mask recognition of the immunogenic β-glucan on the yeast cell wall. While chemotype II requires α-(1,3)-glucan for pathogenesis and virulence, chemotype I isolates still manage to cause disease with an alternative shielding mechanism—a yeast phase cell wall protein encoded by *YPS3* gene to circumvent the need for α-glucan [3, 4].

Recognition of β-glucan by Dectin-1 receptor is detrimental for fungal survival in hosts as the production of essential cytokines that aid fungal clearance, tumor necrosis factor (TNF-α), for example, depends on the activation of this signaling cascade [5]. The engagement between β-glucan and Dectin-1 can be thwarted by the cell wall protein Eng1, a gluconase that hydrolyzes β-(1,3)-glycosyl linkages. It is present on both G186A and G217B strains and acts to degrade β-glucan, thus weakening the interaction between this carbohydrate and its cognate receptor [6]. Despite the shared masking strategy of these two strains, the difference in α-(1,3)-glucan contributes to divergent virulence. Surprisingly, G217B (α-glucan^{-}) is typically considered to be more virulent than G186A (α-glucan^{+}). This finding is true for high inocula of the fungus. However, at low inocula, the differences between these two fungal strains are minimized [7]. Since the chemotypic difference becomes insignificant at low exposure, immunocompromised individuals are likely to be equally susceptible to each chemotype if the inoculum is low. Investigating the pathogenesis in immune-deficient models with a variety of *H. capsulatum* strains may offer valuable clinical insights for disease control.

Adaptation and Growth

The permissibility of the phagocytes, impacted by their activation status, is the decisive point in determining whether the infection will progress. The ingestion of yeasts by activated phagocytes triggers a superoxide burst as an initial attempt to kill. In order to survive, this fungal pathogen secretes enzymatic and nonenzymatic molecules to neutralize the oxidative species. In addition to SOD3, an extracellular superoxide dismutase required for full virulence, two catalases decrease reactive oxygen species (ROS)-mediated damage [8, 9]. The peroxide stress is alleviated by either one of these catalases; only the loss of both extracellular CatB and intracellular CatP results in a reduction of *H. capsulatum* virulence in vivo. The combined deletion of SOD3 and CatB does not further increase the sensitivity of yeast cells to oxidation, suggesting a linear pathway of $O_2^{-} \rightarrow H_2O_2 \rightarrow H_2O$ detoxification [9].

As a phagosome matures, it fuses with a lysosome to form a phagolysosome. The neutral environment within the phagolysosome acidifies which activates lysosomal hydrolases to enzymatically lyse the microbes. Attenuating acidification is an important defense mechanism of *H. capsulatum* to facilitate iron acquisition without activating hydrolases. The fungus alkalinizes the phagolysosome to a pH of ~6.3. This change blunts the biological activity of hydrolases yet permits yeast cells to acquire iron from transferrin [10]. Mutant strains incapable of lysing macrophages (Mφ) reveal a requirement for 3-hydroxy-3-methyl-glutaryl coenzyme A lyase (HCL1) to catabolize leucine and maintain a pH of 6.0–6.5 in lysosomes. The mutation in *HCL1* impairs phagolysosomal acidification resulting from accumulation of leucine intermediates, hence compromising growth of yeasts in Mφ and reducing strain virulence. HCL1 also contributes to utilizing leucine as a carbon source in the glucose-poor environment of phagosomes in vivo, indicating a dual function of this enzyme for yeast survival—combating damage and nutrient limitation [11]. In human phagocytes, acidification to pH of 4–5 is not required for activation of lysosomal enzymes to confer their digestive activity contrary to murine Mφ [12]. Notably, the hydrolases can function at near neutral pH as the heat-killed yeast is degraded at pH of ~6.5; however, the phagolysosome fusion in human Mφ is prohibited which obstructs the delivery of those lysosomal enzymes to the phagosome. Given the disparity between mouse and human, it is crucial to conduct research in human system for mechanistic validation.

Temperature prompts conversion of conidia or mycelial fragments to budding yeast. This transformation is associated with production of essential proteins for cell wall remodeling, growth, infectivity, and pathogenesis [13]. Conidia can be directly engulfed by phagocytes before conversion, and a dichotomous fate of spore is dictated by cell type. Dendritic cells (DC) restrict conidia to yeast transformation; in comparison, alveolar Mφ (AMφ) allow this conversion and are permissive to yeast replication [14]. The parasitic yeast form is critical to evade phagocytic killing. Yeasts use Mφ as a vessel to deliver them to multiple organs. Those yeast-bearing Mφ can become apoptotic both in vivo and in vitro [15]. One of the triggers for apoptosis and cell lysis is the formation of a "crown" structure by yeast aggregates. This occurs as soon as 5 h after in vitro infection and is coupled with DNA fragmentation of Mφ [16]. In addition, calcium-binding protein 1 (CBP1) secreted by *H. capsulatum* activates caspase-3/7-dependent apoptosis [17]. The lack of this protein compromises the virulence of G217B and highlights an active measure by this intracellular fungal pathogen to survive and disseminate through autonomously inducing apoptosis and cell lysis.

The adaptation to the hypoxic environment also influences fungal growth. The compact structure of granulomas formed during infection creates a hypoxic environment to yeasts. *H. capsulatum* is capable of remaining viable under such condition by upregulating Srb1, a homolog of human sterol regulatory element-binding proteins (SREBPs) responsible for sterol synthesis [18]. The anaerobic environment limits ergosterol synthesis and compromises the ability of yeasts to divide. Therefore, the induction of SREBPs becomes key to circumvent the requirement of oxygen for sterol biosynthesis [19].

Zinc is an abundant metal in organisms that serves as a nutrient for growth as well as a cofactor of many signaling molecules and enzymes. Like other fungal species, *H. capsulatum* adopts a zinc sequestration mechanism to maintain zinc homeostasis under low zinc environment. A putative zinc transporter *Hc*Zrt2 identified in *H. capsulatum* is an

orthologue of Zrt1 in *Saccharomyces cerevisiae*. Mice infected with *Hc*Zrt2 mutant strain withstand lethal infection, suggesting this zinc transporter is necessary for the virulence of the organism [20]. Clearly, *H. capsulatum* employs numerous measures to quickly adapt to environmental stress. The linear and lateral cooperation of different molecules or signals maximizes the chance of this fungus to sustain. Further investigation is warranted regarding the interplay between different mechanisms for evading destruction by host cells.

Pathogenesis From Perspective of Host Immunity

Innate Immunity

Efficacious adaptive immunity requires an appropriate innate response [21, 22]. Mφ clearance of *H. capsulatum* hinges on recognition by pattern recognition receptors and stimulation by proinflammatory cytokines, both which initiate signaling deleterious for the fungus [23, 24]. Failure to generate appropriate levels of necessary cytokines, including interferon γ (IFNγ) or granulocyte macrophage colony-stimulating factor (GM-CSF), or overproduction of the type 2 cytokine interleukin (IL)-4 or anti-inflammatory cytokine IL-10 sways Mφ to an alternatively activated phenotype and dampens clearance of *H. capsulatum* [25–27] (Fig. 1b). A collapse in the innate immunity leads to persistent infection and can evolve into disseminated disease and death.

Lipid Mediators

Lipid mediators play divergent roles in inflammation by modulating cell recruitment and altering phagocytosis and killing by Mφ [28–33]. Airway epithelial cells and inflammatory cells generate these lipids, including prostaglandins (PG), leukotrienes (LT), and platelet-activating factor (PAF). The enzyme 5-lipoxygenase (5-LO) synthesizes LT from arachidonic acid and deletion of 5-LO causes mice to succumb to *H. capsulatum* by 20 days post infection (p.i.) [34]. LTB_4 and PAF recruit neutrophils (PMN) to the primary site of infection [35]. Both human and murine PMN exhibit fungistatic activity against *H. capsulatum* and promote *H. capsulatum* clearance [36, 37]. During experimental histoplasmosis, cells in the lungs express LTB_4 and LTC_4 beginning at day 1 p.i. that contributes to PMN recruitment [28]. Directing PMN is a dynamic process of the host in that invasion of PMN in a remote site by exogenous administration of LTB_4 and PAF is subverted upon infection. This indicates a surge of endogenous LTB_4 and PAF elicited by pulmonary challenge of *H. capsulatum* draws those PMN to the infection site. While inhibition of LTB_4 receptor or PAF receptor alone partially decreases PMN migration, concomitant blockade hinders PMN influx to both lungs and remote site, which indicates cooperation between the two. 5-LO knockout mice were utilized to investigate the role of cross-talk between LT and PAF. In the absence of endogenous LT in 5-LO knockouts, exogenous administration of PAF is not capable of recruiting PMNs. This suggests the chemotactic properties of PAF are dependent on 5-LO activation, specifically the presence of LT [38]. One caveat in interpreting these data is that the impact of PMN in overcoming *H. capsulatum* infection is not fully understood. Depletion of PMN by Gr-1 monoclonal antibody has been used to indicate their importance in host defenses. However, this antibody eliminates inflammatory monocytes (iMo) as well. In studies which use a more specific antibody (directed against the surface antigen Ly6-G), fungal burden is not altered at 7 days p.i. [39–41].

As opposed to LTB_4 and PAF promoting inflammation, PGs suppress the inflammatory response. Cyclooxygenase (COX)-1 and -2 produce PGE_2 which enhances IL-10, IL-4, and IL-5 generation to induce Th2 responses [31, 42, 43]. PGE_2 also hinders the synthesis of IL-2 and IFNγ that dampens Th1 response and prevents Mφ phagocytosis and bacterial killing [31–33, 44]. Loss of PGE_2 by blocking COX-2 with celecoxib in mice reduces the fungal burden in the lungs and spleen through increasing nitric oxide, LTB_4, and INFγ. COX-2 inhibition improved Mφ activation and phagocytosis and prolonged survival of mice infected with a lethal dose of *H. capsulatum* [45]. The two studies reveal the importance of coordination of pro- and anti-inflammatory lipid mediators in controlling fungal infection.

Cytokine Regulation

Modulating the influx of immune cells into the lungs by chemokines to initiate a Th1-type response is paramount in combating *H. capsulatum* infection [27]. Not only does CCR2 instigate migration of monocytes from the bone marrow into the blood and then tissue, it also promotes a Th1 response and controls pathogens [46–52]. CCR2 knockout mice display enhanced susceptibility to *H. capsulatum* infection with decreased monocytes and DC and higher quantities of IL-4 in lungs beginning at day 3 p.i. compared to wild type (WT) controls [26]. IL-4 stimulation of Mφ induces an alternatively activated phenotype; this produces a permissive environment for *H. capsulatum* growth possibly by creating a zinc-rich environment and/or blunting GM-CSF signaling [53].

Detection of upregulated IL-4 as early as day 3 of infection suggests an innate cell source of this cytokine rather than an antigen-reactive T cell since adaptive immunity is not activated until day 7. Although innate lymphoid cells (ILC) type 2

would be the likely origin of initial IL-4 [54], in CCR2$^{-/-}$ mice, eosinophils are the source of the detrimental overproduction of IL-4. Their importance in cytokine generation is supported by finding that depleting them in *H. capsulatum* infected CCR2$^{-/-}$ mice diminishes fungal burden. Likewise, IL-5 overexpressing mice that are hypereosinophilic manifest amplified IL-4 and compromised host resistance to the fungus. Release of IL-4 by eosinophils requires CR3-dependent phagocytosis and Syk kinase signaling [55]. The genesis of IL-4 in CCR2$^{-/-}$ mice is not the final cause of failure to control the pathogen. IL-4 in CCR2$^{-/-}$ mice elicits amplified IL-33 production by lung Mφ and not epithelial cells. This finding stands in contrast to the vast majority of data that IL-33 drives the generation of IL-4 [56]. Together, *H. capsulatum* and IL-4 work synergistically to induce IL-33, which alternatively activates Mφ and impairs host response to *H. capsulatum* infection [57]. Eosinophils and IL-4 cause pathogenesis in allergic diseases prompting the question if allergies and asthma heighten susceptibility to histoplasmosis.

After the recruitment of immune cells to the lungs, activation of iMo and Mφ by GM-CSF is necessary to clear the fungus [58]. GM-CSF activates both human and murine Mφ, while stimulation with IFNγ only inhibits fungal growth in murine Mφ in vitro [24]. One mechanism by which GM-CSF exerts antifungal activity on Mφ is through zinc appropriation [53]. A deficiency in GM-CSF results in death in *H. capsulatum* infection [58]. Activation of Mφ by GM-CSF causes the Mφ to preferentially sequester zinc by inducing zinc-binding metallothioneins and upregulating the zinc transporters *Slc30a4* and *Slc30a7*. Mφ seizure of zinc leads to enhanced ROS production and also prevents the yeast from utilizing the metal [59]. This work highlights the mechanism for GM-CSF-activated Mφ killing of *H. capsulatum* through zinc sequestration.

Classical activation of Mφ necessary to combat infection is controlled by many transcription factors including NFκB, STAT family, IRF family, and AP-1 [60]. Hypoxia-inducible factor 1α (HIF-1α), a key transcription factor in response to hypoxia, regulates numerous genes involved in innate and adaptive immune responses. HIF-1α directly targets TNFα, IL-12, IL-10, and CCL2 in myeloid cells [61–64]. Compared with WT control, myeloid HIF1-α-deficient mice exhibit increased fungal burden beginning at day 3 p.i. and decreased survival. The dampened immune response is associated with a surge in IL-10 synthesis largely by Mφ in the lungs, which depends on CREB-binding protein [41]. This demonstrates a role for HIF-1α in Mφ regulation beyond reaction to hypoxia.

Innate–Adaptive Coordination

The coordination of phagocytosis and subsequent T cell activation by Mφ and DC is pivotal to the initiation of an adaptive immune response [22]. IFNγ, TNFα, IL-12, IL-1β, CCL2, and GM-CSF are produced to recruit and activate both innate and adaptive cells [26, 65–67]. Myeloid differentiation primary response gene 88 (MyD88) is a central adaptor protein in several immune signaling pathways, including Toll-like receptors (TLRs) and cytokine signaling through IL-1R and IL-18R. Disrupting MyD88 in other fungal disease models results in defective innate and adaptive immunity [68–71]. In experimental histoplasmosis, MyD88 knockout mice display decreased IL-6, IL-1β, IL-12, CCL2, and CXCL1 production by AMφ and DC that causes diminished iMo recruitment to lungs. This defect in cytokine production and cellular recruitment impedes T cell activation and leads to enhanced fungal burden and death [72]. The mechanism for innate cell recognition of *H. capsulatum* is not fully elucidated; this work may suggest a role for TLRs.

Another factor important in driving inflammation during *H. capsulatum* infection is the galectin family protein galectin-3 (Gal3), which is expressed in Mφ, DC, lymphocytes, and epithelial cells [73]. Gal3 plays a diverse role in immunity; it attracts PMN and deletion of Gal3 reduces PMN recruitment in *Streptococcus pneumoniae* and *Toxoplasma gondii* infections [74, 75]. It also regulates T helper responses in infection, autoimmune disease, and allergy [76–79]. DC from Gal3 knockout mice infected with *H. capsulatum* engender higher levels of IL-23, TGFβ1, and IL-1β that are necessary to stimulate IL-17 production in both PMN and T cells. Greater IL-17A and reduced IFNγ in the lungs shifted the balance from a Th1 to a Th17 response [80]. This further reveals the crucial intersection of innate and adaptive coordination.

Adaptive Immunity

Protective Response

Similar to combating other intracellular pathogens, mounting a dominant Th1 response is vital for *H. capsulatum* clearance. In addition, an induction of IL-17 producing T cells contributes to optimal inflammatory and protective responses [67, 81]. IL-17 compensates but does not replace the function of IFNγ when Th1 is impeded and thus dispensable for host resistance [72, 82]. In contrast, efficient vaccine immunity against endemic mycosis depends on IL-17 producing T cells. Both CD4$^+$ and CD8$^+$ T cells give rise to this cytokine upon vaccination. The differentiation of Th17 cells requires innate cells to sense Dectin-1 and Dectin-2 via binding to β-glucan [82]. In contrast, the differentiation of IL-17 producing cytotoxic T cells (Tc17) is independent of Dectin-1 pathway. These cells confer vaccine-mediated immunity in mice that lack CD4$^+$ T cells. The impairment in IFNγ producing CD8$^+$ T cells (Tc1) due to the loss of IL-12 signaling does not alter Tc17 responses or vaccine derived resistance [83]. These results indicate that vaccination for the endemic mycoses can be effective in those with absent or defective CD4$^+$ T cells.

Non-protective Response

Th2 and Treg dampen the immune responses against infection with *H. capsulatum*. Several studies affirm that an elevation in IL-4 retards fungal resolution [25, 26, 84]. DC are the most important antigen presenting cells that control the polarity and magnitude of Th2. A Krüppel-like factor family transcription factor KLF2 previously found to be important for licensing cell activation negatively regulates IL-4 production by $CD4^+$ T cells via Jagged2/Notch pathway; deletion of this factor in myeloid cells, specifically DC, causes deleterious fungal resolution as a result of IL-4 elevation [84]. Treg expansion is also connected with DC modulation during fungal infection [85]. Anti-TNFα treatment elevates Treg in mice infected with *H. capsulatum* [86]. Though the cause for Treg induction remains unresolved, it is conceivable that tolerogenic DC may contribute to amplifying this population and increase susceptibility. The regulation of DC shapes the adaptive immune response, dictates the course of disease, and is suggested as a promising vaccination strategy for lethal fungal infections [87]. In short, elevating Th1/17 and inhibiting Th2/Treg would be a useful tool to aid fungal resolution.

Risk Factors

Disseminated histoplasmosis is associated with compromised immunity as a result of disease and/or treatment. Patients with HIV, hepatitis, or other fungal diseases are more likely to succumb to infection if not properly diagnosed and treated. The inhibition of immune functions by anti-TNFα or corticosteroids for inflammatory diseases/organ transplantation also greatly increases the susceptibility for disseminated histoplasmosis.

Recent case reports described two gene mutations as risk factors for different degrees of histoplasmosis. Patients with STAT1 gain-of-function mutation are prone to develop disseminated infection. This is likely attributed to dysregulated inflammation caused by IFNγ tachyphylaxis [88]. To a less severe extent, patients with STAT3-mutated hyper-IgE syndrome are predisposed to extrapulmonary histoplasmosis in endemic regions. Loss-of-function of STAT3 could attenuate Th17 responses and a suboptimal defense [89].

New Diagnostic Targets and Treatment for Histoplasmosis

Testing surface antigens is a fast and sensitive measure for disseminated or pulmonary histoplasmosis following high exposure [90]. Two novel *H. capsulatum* surface antigens that are not associated with virulence are suggested as a diagnostic tool for serological test of active *H. capsulatum* infection [91, 92]. Culture filtrate protein 4 and *N*-acetylated α-linked acidic dipeptidase can be detected using ELISA. The latter has also been validated in real-time PCR assay [93]. Given the immunoassay for testing *H. capsulatum* antigens is considerably more sensitive and less invasive in urine than sera, assessing the presence of these two antigens in urine samples from patients is highly valuable for future application [94–96].

The narrow selection of anti-fungal drugs calls for research and development of new drugs and treatment options. This is extremely important for severe and disseminated histoplasmosis given the high mortality rate and the nephrotoxicity of the first-line drug amphotericin B. Combination therapy employing farnesol as an adjuvant significantly reduces the minimal inhibitory concentration of amphotericin B, azoles, and caspofungin. Among these combinations, intraconazole and farnesol produce the strongest anti-fungal effect in *H. capsulatum* yeast, mycelial, and biofilm [97, 98]. A formation of biofilm in many fungal species is associated with increased virulence which imposes the need for drugs that can effectively penetrate and kill [99]. Though *H. capsulatum* is capable of producing biofilms with in vitro studies, there is no evidence that *H. capsulatum* forms biofilms in vivo or that these structures alter antifungal susceptibility.

Aside from new azole drugs such as isavuconazole and aminothiazole that were recently evaluated, other classes of drugs for treating mild to moderate histoplasmosis have been studied as additional options for treatment [100–102]. Hydrazones, derived from isoniazid which is a primary drug in treating tuberculosis that shows anti-fungal activities, can impair ergosterol synthesis and membrane integrity, as well as inhibit mature fungal biofilm [103, 104]. Miltefosine and levamisole, previously used for cancer and helminth treatment respectively, confer effective fungistatic activity via regulating ergosterol synthesis and membrane permeability [105]. Immunomodulatory strategies that target host immune responses were also explored, exemplified by a pharmacological COX-2 inhibitor celecoxib and monoclonal antibody against heat shock protein 60 [45, 106]. These new promising therapeutic measures warrant further validation in clinical settings.

Conclusion

H. capsulatum evades the host response by numerous mechanisms including incorporating unrecognizable glycoproteins into the cell wall, secreting molecules that neutralize oxidative species, and inducing host cell lysis. To combat infection, early recruitment and response of PMN and activation of Mφ in *H. capsulatum* infection are required for mounting an appropriate adaptive immune response and controlling infection. Th1/Th17 response confers optimal control of fungal growth, whereas overproduction of Th2 cytokines leads to vulnerability to *H. capsulatum* infection. The antigen

presenting cells serve as a bridge between innate and adaptive immunity to regulate the bias and strength of responses weighing upon the pathogen burden.

The combination of drugs that target biological processes of *H. capsulatum* for survival and immune responses is likely to synergize to combat virulent infection with low risk of toxicity. The aforementioned strategies could potentially improve patients' overall health and survival. It is of great importance to continue expanding the pool of anti-fungal therapies in light of continuing research of pathogen, host immunity, and their interactions.

Compliance with Ethical Standards

Conflict of Interest Jamie L. Tweedle, Ye Xiong, and George S. Deepe Jr declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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