Diagnosis of Pulmonary Histoplasmosis

Subjects: Microbiology

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Acute pulmonary histoplasmosis (APH) typically presents with fever, chills, shortness of breath, and resembles community acquired pneumonia. APH can range from a mild self-limiting illness to acute respiratory distress syndrome. Subacute pulmonary histoplasmosis (SPH) has a more insidious onset over at least one month and may develop after a smaller inoculum exposure. Chronic pulmonary histoplasmosis (CPH) is classically seen in older males with underlying lung disease. CPH has a similar presentation to tuberculosis with fever, night sweats, weight loss, cough, and dyspnea over at least three months. H capsulatum may also cause pulmonary nodules, mediastinal adenitis, mediastinal granulomas, and mediastinal fibrosis. Progressive disseminated histoplasmosis is a form of histoplasmosis that result from hematogenous spread and can impact multiple organ symptoms including the respiratory tract and cause severe disease.

Keywords: Histoplasmosis

1. Histoplasmosis

Histoplasmosis is caused by *Histoplasma capsulatum var capsulatum* and *Histoplasma capsulatum var. duboisii* [1]. Classically, *H capsulatum* is thought of as endemic to the Ohio and Mississippi River Valleys in the United States, as well as parts of Central and South America [2][1][3][4][5]. More recently it has become clear that *Histoplasma* occurs frequently in many parts of the world including: Central and Eastern North America, the majority of Central and South America, much of sub-Saharan Africa, large portions of southeast Asia and small areas within Australia and Europe [1]. *H duboisii* has primarily been described in West Africa.

Infection is acquired through inhalation of spores from soil that is contaminated with bird or bat droppings [4][5]. *Histoplasma* can cause a wide variety of clinical manifestations including a spectrum of pulmonary diseases ranging from acute to chronic presentations [4][6].

2. Symptoms

Acute pulmonary histoplasmosis (APH) typically presents with fever, chills, shortness of breath, and resembles community acquired pneumonia [2][7][4][6][8]. APH can range from a mild self-limiting illness to acute respiratory distress syndrome [2][4][6][9]. Subacute pulmonary histoplasmosis (SPH) has a more insidious onset over at least one month and may develop after a smaller inoculum exposure [2][7][4][6]. Chronic pulmonary histoplasmosis (CPH) is classically seen in older males with underlying lung disease [2][7][5][6]. CPH has a similar presentation to tuberculosis with fever, night sweats, weight loss, cough, and dyspnea over at least three months [4][5][6][8]. *H capsulatum* may also cause pulmonary nodules, mediastinal adenitis, mediastinal granulomas, and mediastinal fibrosis [10][2]. Progressive disseminated histoplasmosis is a form of histoplasmosis that result from hematogenous spread and can impact multiple organ symptoms including the respiratory tract and cause severe disease [6].

In APH, imaging frequently shows diffuse bilateral patchy opacities with hilar and mediastinal adenopathy while diffuse reticulonodular or miliary infiltrates can be seen less commonly [2][7][6][9][11]. In CPH patchy infiltrates can progress to large, destructive cavities; hilar and mediastinal lymphadenopathy are uncommon compared to APH [5][6].

3. Diagnosis

Identification of *H capsulatum* on histopathology and culture is the classical diagnostic standard [2][5][12]. The narrow based budding ovoid *Histoplasma* yeast (2–4 μM in diameter) is visualized via direct microscopic examination or the use of Gomori methenamine silver, Giemsa, periodic acid-Schiff, or hematoxylin eosin stains of specimens such as respiratory samples, lymph node tissue, or lung tissue (Figure 1A) [10][2][3][4][6][13][14][15]. Cytopathologic examination of bronchoalveolar lavage (BAL) fluid is positive in up to 50% of cases [2][7][3]. Histopathologic examination can reveal both caseating and non-caseating granulomas [3][4]. Despite the potential to improve diagnosis, pathological examination is not

feasible in most patients as it requires invasive procedures, such as bronchoscopy or biopsies [3]. In general, it is more useful in disseminated histoplasmosis compared to pulmonary histoplasmosis and is more likely to be positive in SPH or CPH compared to APH [6][16]. *Histoplasma* can take 2–8 weeks to grow on culture which is similarly more likely to be positive in SPH or CPH compared to APH [2][7][3][4][5][13][16]. Overall sensitivity of culture of sputum or bronchoscopy specimens is 48–75% in pulmonary histoplasmosis [7][6].

Antigen detection can provide rapid diagnosis of pulmonary histoplasmosis. Numerous commercial and in-house tests are available, however, agreement between tests is not uniform [8][12][17]. Most though not all antigen tests use an enzyme immunoassay (EIA) [8][12][18]. Antigen is generally more likely to be positive in APH compared with SPH and CPH although CPH commonly yields positive results as well [2][7][6][16]. In a multicenter evaluation by Hage et al., antigenuria was detected in 83% of acute cases, 30% of subacute cases, and 87.5% of chronic pulmonary histoplasmosis, with the highest antigen concentrations in acute cases—combined urine and serum antigen testing improved yield [16]. In another large study of APH, antigen was detected in serum and urine in 65% and 69% of cases, respectively [9]. This same study found that antigen was more likely to be detected in patients who require hospitalization, likely reflecting higher fungal burden in more severe disease [9]. Antigen testing of BAL can further aid in diagnosis of pulmonary histoplasmosis, particularly in CPH or diffuse pulmonary disease complicating disseminated histoplasmosis. Hage et al. found that among 31 patients with histoplasmosis and pulmonary involvement, antigen detection in BAL had 93.5% sensitivity, 97.8% specificity, 68.8% positive predictive value, and 99.6% negative predictive value [19]. Overall, 21 of the 31 patients in this study were immunocompromised with disseminated histoplasmosis including disseminated pulmonary disease [19]. One limitation of *Histoplasma* antigen testing is its cross reactivity with other mycoses, such as *Blastomyces* spp, *Talaromyces marneffei*, *Paracoccidioides*, *Coccidioides*, and *Aspergillus* spp. [2][7][5][6][8][11][12][16][18][20][17]

The IMMY *Histoplasma* EIA, is a commercially available, FDA approved EIA for detection of *Histoplasma* antigen in urine [8][12]. In a study by Theel et al. the IMMY EIA had 97.6% agreement with MiraVista Diagnostic's EIA and a specificity and sensitivity of 99.8% and 64.5%, respectively [12]. As opposed to MiraVista's EIA which is done at a central laboratory, health centers can perform IMMY's test given its FDA approval. MiraVista recently developed a lateral flow assay (LFA) for serum antigen detection [21]. In patients with HIV and disseminated histoplasmosis, sensitivity was 96% and specificity 94% [21]. This test is not FDA approved.

Antibody testing is also used to diagnose histoplasmosis. Because antibodies take time to develop after acute infection, they are more useful in SPH and CPH than APH and negative initial antibody testing should be repeated in one to two months if suspicion is high [2][7][3][4][5][6][9][11][14]. Additionally, antibody testing may be negative in immunocompromised patients and may cross react with other endemic mycoses such as *Blastomyces*, *Paracoccidioides*, and *Coccidioides* [2][3][4][6]. Common methodologies include immunodiffusion (ID), complement fixation (CF), or EIA [2][3][4][6]. ID detects H and M bands, H bands are more rare but when found indicate acute infection whereas M bands are more common and may persist for years [2][3][6]. A fourfold rise in CF titers or a single titer of 1:32 or higher is indicative of active infection [2][3][6]. Compared to CF, ID is slightly more specific and less sensitive [5]. One multicenter evaluation found 67% seropositivity (by CF or ID) in APH, compared to 95% in SPH and 83% in CPH [16]. In one study of patients with APH, MiraVista Diagnostics' IgM, and IgG EIA exhibited 89% sensitivity and 92% specificity [11]. Combining antigen and antibody testing may improve sensitivity for diagnosis of APH, potentially as high as 96% [9][11]. Similar sensitivity has been found using the combination of BAL antigen detection and BAL cytopathology [7].

Nucleic acid amplification tests (NAAT) such as polymerase chain reaction (PCR) or loop-mediated isothermal amplification (LAMP) have been utilized for the identification of *H. capsulatum*, however these have variable sensitivities and none are commercially available [2][22][23][24][25][26][27][28][29]. NAATs are less likely to have false positive results due to other endemic fungi compared to antigen and antibody testing [22]. A reference database has been created to identify *H. capsulatum* via matrix assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS) but little data on performance is available [30][31]. Similarly, while metagenomic next generation sequencing (mNGS) has been used on BAL fluid to diagnose *H. capsulatum* causing chronic progressive pulmonary lesions and epiglottis lesions, broader performance data are not available [32][33]. Finally, there may be a role for panfungal PCR to diagnose histoplasmosis, but so far use has been exploratory [34].

Pulmonary infection is more common due to *H. capuslatum var capsulatum* than *H. capsulatum var. duboisii* [1]. In a summary of 94 reported cases, only 10 were suspected to have pulmonary involvement [35]. *H capsulatum var. duboisii* frequently causes lymphadenopathy, bone, cutaneous, sub-cutaneous, and disseminated disease, and may occur decades after leaving the endemic area [35]. *H capsulatum var. duboisii* diagnosis has not been well studied and so diagnostic test performance characteristics are less well understood. Histopathology, cytology are commonly used while

confirmation with culture or PCR are less common and antibody testing is even more rare [35]. Histoplasma antigen testing has not been utilized [35].

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