Histoplasmosis in Eastern Uttar Pradesh: Our five years experience at a tertiary care center

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Abstract:

Histoplasmosis is an endemic fungal infection which primarily infects the immunocompetent as well as immunocompromised patients. It primarily affects the respiratory tract and disseminates hematogenously where it involves the bone marrow, lymph nodes and the adrenal glands. Clinical manifestations of histoplasmosis are of three main types: Acute primary, chronic cavitatory and progressive disseminated histoplasmosis. We are presenting case series of six Histoplasmosis cases, in which *Histoplasma capsulatum* strains were isolated from the bone marrow, lymph node and skin biopsy respectively whereas three cases were diagnosed based on bone marrow examination and histopathological examination and special fungal staining of relevant tissues.

Key words: Bone marrow, *Histoplasma capsulatum,* Progressive disseminated histoplasmosis (PDH) and Pyrexia of unknown origin (PUO)

Introduction:

Histoplasmosis is a systemic fungal disease caused by *Histoplasma capsulatum*, a dimorphic and ubiquitous fungus. The disease is endemic in certain areas of North, Central, and South America,

as well as Africa and Asia [1]. Most primary infections with *H. capsulatum* are either asymptomatic or result in mild influenza-like illness; however, certain forms of histoplasmosis can cause life-threatening infections with considerable morbidity [2, 3]. The natural habitat of this fungus is soil that has been contaminated with bird or bat droppings. Pulmonary infection usually develops through inhalation followed by haematogenous spread to the reticuloendothelial system within a few weeks [4, 5].

Highly infectious soil is found to be source of infections caused by *Histoplasma capsulatum*. Most of the time infection remains asymptomatic. Those who develop clinical manifestations usually are immunocompromised or are exposed to a high quantity of inoculum. The extent of disease depends on the number of conidia inhaled and the function of the host's cellular immune system. Pulmonary infection is the primary manifestation of histoplasmosis, varying from mild pneumonitis to severe acute respiratory distress syndrome. In those with chronic lung disease a chronic progressive form of histoplasmosis can occur. Dissemination of *H. capsulatum* within macrophages is common and becomes symptomatic primarily in patients with defects in cellular immunity.

Case details:

In all suspected cases of histoplasmosis, a diagnosis was achieved by isolation of *Histoplasma capsulatum* by fungal culture, or visualization of appropriate morphologic fungal forms in the relevant tissues or bone marrow aspiration. Culture remains the gold standard for its diagnosis, but it requires a lengthy incubation period two to four weeks (Fig.1and2) Fungal staining produces quicker results than culture but is less sensitive [4]. Culture for acid fast bacilli was also found to be negative. Serological test for *Leishmania* was negative. Fungal culture was not done in all cases. Bone marrow aspirate stained by Leishman stains showed oval globose yeast like cells measuring

3-4 mm x 2-3 mm in size, suggestive of *Histoplasma capsulatum*(Fig.3,4 and 5). PAS stained revealed these yeast-like cells as bright eosinophilic structures. Direct examination of smear relating to clinical symptoms helped in provisional diagnosis, Isolation of organisms by culture was not successful but histomorphological features were characteristic for diagnosis in two cases. Salient patient details are given in Table 1.

Table.1 Clinical & Laboratory profile of patients

	Age/ sex	**Clinical Manifestations**	**Retro Positive**	**Organ Involved**	**Diagnosis by Direct Microscopy**	**Culture**	**Management**	**Outcome**
	50/ M	Fever with hepatosplenomegaly with skin rash	No	Liver, spleen and bone marrow	Yeast cell seen	Sample was not sent for Microbiological work up.	Inj. Amphotericin B and Itraconazole 200mg BD	Survived
	35/ M	Enlarged axillary lymph nodes	Yes	Lymph nodes	Negative	On culture at 25^0C, there was cotton white growth. On LCB wet mount, numerous tuberculate macroconidia and few microconidia	Inj. Amphotericin B and Itraconazole 200mg BD	Survived
	40/F	PUO intermittent high-grade	Yes	Bone marrow	Yeast cells seen	On culture at 25^0C, there was cotton white growth.	Inj. Amphotericin B and	Died

		fever with weight loss				On LCB wet mount, numerous tuberculate macroconidia and few micro conidia	Itraconazole 200mg BD	
	67/M	PUO with abdominal pain	No	Adrenal glands	Intracellular yeast cells	Sample was not sent for Microbiological work up.	Inj. Amphotericin B and Itraconazole 200mg BD	Died
	56/M	PUO with weight loss, skin lesions	No	Skin lesions	Intracellular yeast cells	On culture at 25^0C, there was cotton white growth. On LCB wet mount, numerous tuberculate macroconidia and few micro conidia	Inj. Amphotericin B and Itraconazole 200mg BD	Improved
	46/M	Abdominal pain and darkening of the skin	No	Adrenal glands	Intracellular yeast cells	No growth	Inj. Amphotericin B and Itraconazole 200mg BD	Improved

Discussion:

Histoplasmosis is a systemic fungal disease acquired by the inhalation of microconidia of the filamentous phase of the fungus *Histoplasma capsulatum.* The severity of clinical manifestation depends on size of inoculum, underlying health of the person and immune status to *Histoplasma capsulatum*. Immunocompetent patients present as limited respiratory infection comprising of fever, malaise, cough and chest pain [6, 7, and 8]. These are non-specific symptoms that may also

occur with multiple other diseases. The majority of affected individuals remain clinically silent and display no apparent symptoms [5, 9] Most of our patients were middle aged who had shown significant hepatosplenomegaly with bone marrow involvement. Chronic infection tends to present in older, immunocompetent patients as pancytopenia, hepatosplenomegaly, oropharyngeal and/or skin lesions, gastrointestinal involvement, and signs and symptoms of adrenal gland dysfunction. Disseminated histoplasmosis may present as acute PDH with fever, malaise cough mimicking pulmonary tuberculosis. Chronic PDH presents as fever, sweats, weight loss, organomegaly and lymphadenopathy. Another possible explanation of underdiagnosed is that disseminated histoplasmosis resembles visceral leishmaniasis in many aspects with feature of fever, weight loss and hepatosplenomegaly. Moreover, both of these are responsive to Amphotericin B. Diagnostic accuracy has improved greatly with the use of an assay for Histoplasma antigen in the urine; serology remains useful for certain forms of histoplasmosis, and culture is the ultimate confirmatory diagnostic test [5]. Classically, histoplasmosis has been treated with long courses of amphotericin B. Itraconazole is the azole of choice following initial amphotericin B treatment [6]. Treatment with fluconazole 200 mg to 400 mg daily appears to be even less effective than ketoconazole and itraconazole [6]. Disseminated histoplasmosis is aggressive, progresses rapidly, and requires early and effective treatment; our patients were treated with amphotericin B deoxycholate antifungal infection and itraconazole. The majority of these patients were successfully treated, whereas two of cases died.

Conclusion

A high need of suspicion will lead to pertinent samples being sent to mycology tests for correct diagnosis. It is thus important for microbiologist to look for dimorphic fungi in fever of unknown

origin. This will not only lead to proper identification of microorganism but will also provide us correct treatment options.

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Fig. 1 Growth on SDA at 25^{0}C

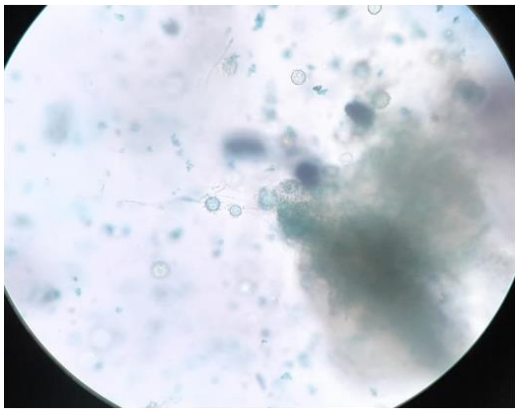

Fig.2 LCB wet mount showing Tuberculate macroconidia

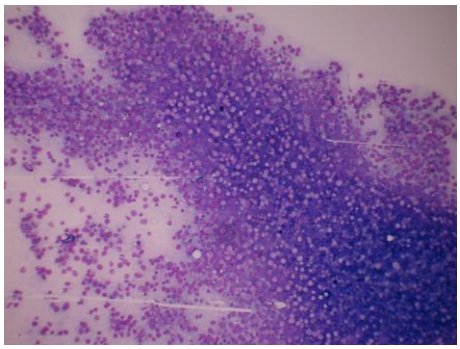

Fig 3. H&E staining of lymph node showing the veast cells.

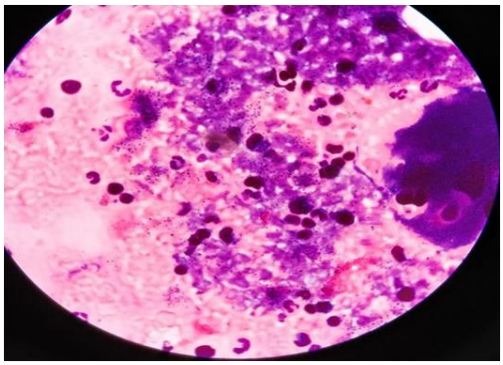

Fig.4 Giemsa staining of Bone marrow showing the yeast cells.

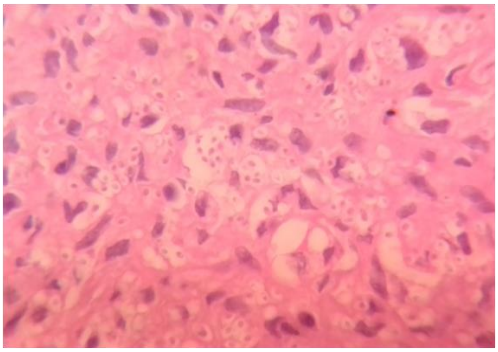

Fig 5. H& E staining of Adrenal gland showing Intracellular yeast cells.