

## **Cryptococcosis: Its Prevalence and Clinical Presentation among Hiv Positive and Negative Patients in Rims, Manipur**

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**Abstract:** *Cryptococcus neoformans*, the causative fungal agent of cryptococcosis remain a common cause of infectious morbidity and mortality, especially among HIV-positive patients living in Sub-Saharan Africa and South-East Asia. This study was undertaken to evaluate the prevalence and clinical presentation of *Cryptococcus* infections among HIV positive and negative patients in RIMS, Manipur. Specimens like CSF, sputum, urine, blood, tissue biopsy or aspirates from clinically suspected cryptococcosis cases from RIMS hospital, were subjected to mycological examination. Out of the 48 patients enrolled for the study, *Cryptococcus* spp were isolated from 16 (33.33%) patients. Among these 16 cryptococcosis patients, majority of them presented with cryptococcal meningitis 13 (81.25%), while 1 (6.25%) patient each presented with cryptococcal lymphadenitis, disseminated cutaneous cryptococcosis and osseous cryptococcosis respectively. Also, of these 16 cryptococcosis patients, 14 (87.5%) were HIV positive. Among these HIV positive, sexual transmission with 64.29 % forms the commonest route of transmission, followed by intravenous drug injection with 35.71%. Out of these 14 HIV positive patients, 11 (78.57%) were on ART. This study shows high prevalence of cryptococcosis in our region with the most common presentation being cryptococcal meningitis. Also, there was high incidence of cryptococcosis among HIV-infected patients.

**Key words:** *Cryptococcosis, Cryptococcus neoformans, HIV*

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### **I. Introduction**

*Cryptococcus neoformans* is an encapsulated yeast, ubiquitously found worldwide in soil contaminated with bird droppings mainly of pigeons, roosting sites and decaying woods and vegetables. The infectious propagules of this fungus from these environments may be the source of human infections [1,2]. *Cryptococcus neoformans*, the causative agent of cryptococcosis, can cause life threatening infections with predilection for brain and meninges, occasionally lungs and skin. Although very rare, it may involve in other organs such as the heart, liver, spleen, kidneys, thyroid, lymphnodes, adrenal glands, eyes, genitourinary tract, gastrointestinal tract and abdominal, bones and joints [3]. The emerging menace of cryptococcosis has assumed global proportion over the years due to climate changes and the worldwide spread of HIV infection as well as the increasing number of patients with impaired immunity as a result of cancer chemotherapy, diabetes mellitus, sarcoidosis, steroids, renal transplant or inherited immune defects. It also causes infection less commonly in immunocompetent hosts [4,5]. Although effective treatment for HIV disease has led to significant decrease in the incidence of cryptococcal meningitis (CM) in the western countries, it continues to be the common cause of infectious morbidity and mortality especially among the HIV-positive patients living in Sub-Saharan Africa and South-East Asia [6,7,8,9].

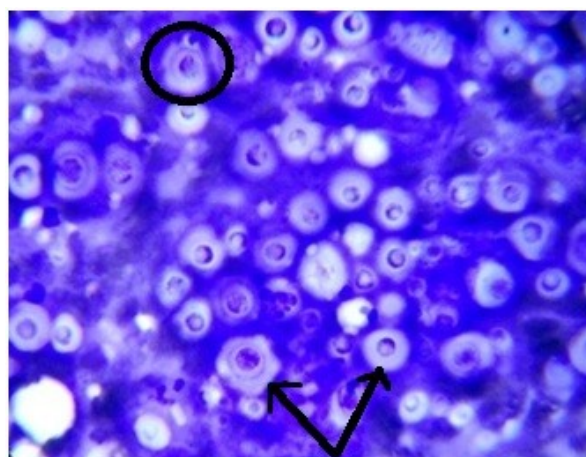
Due to the high prevalence of HIV infection in our state Manipur [10], a large percentage of the population continues to be at risk for infection with indigenous *Cryptococcus* strains. This study was undertaken to evaluate the prevalence and clinical presentation of *Cryptococcus* infection among both the HIV positive and negative patients in Regional Institute of Medical Sciences (RIMS) hospital, Manipur.

### **II. Materials and methods**

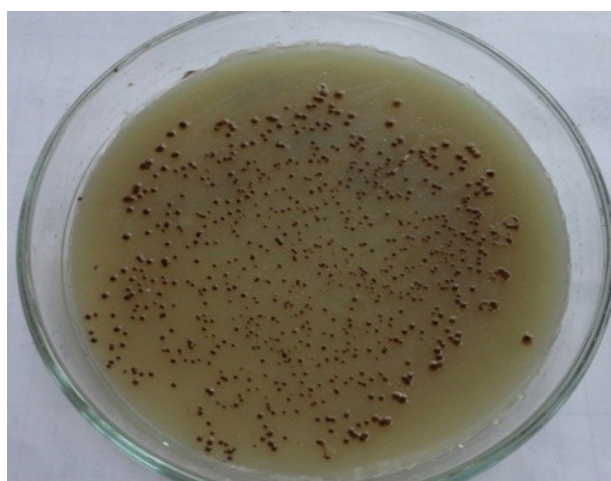
The study was conducted in the Microbiology Department, RIMS, Imphal Manipur, with a study period of one and a half years starting from November 2011 to October 2013. Patients enrolled in the study were taken informed consent and in the case of minor, informed consent were taken from parents. This was a Cross sectional study carried out under the approval of the institutional ethical committee, RIMS.

The study consist of 67 samples, collected from 48 clinically suspected cryptococcosis cases in RIMS hospital and were subjected to mycological examination. The specimens included CSF, sputum, urine, blood, tissue biopsy or aspirates depending upon the site of involvement i.e. from superficial lesions to deep seated infections. Multiple samples were taken from a single patient. The specimens were subjected to examination by India ink preparation, Meyer's mucicarmine stains, Giemsa stains (Fig.1) and antigen detection (CALAS, Meridian Diagnostics, Cincinnati, Ohio). The specimens were further confirmed by fungal culture on Sabouraud

dextrose agar (Emmon's modification with antibiotics chloremphenicol) and laboratory-prepared Bird seed agar plates (BSA) which showed mucoid colonies (Fig.2). Isolates were identified as *C. neoformans* by growth at 37°C, urease test, inositol assimilation, subculture on L-canavanine glycine bromothymol blue (CGB) and creatinine dextrose bromothymol blue thymine (CDBT) medium detected by color reaction. The identity of the isolates were also confirmed by automated Vitek 2 yeast identification system [bioMerieux Inc., (Durham, NC) USA]. The datas so collected were processed using SPSS version 16.0.



**Figure 1:** Encapsulated *Cryptococcus* in skin aspirate's Giemsa stain(x1000)



**Figure 2:** BSA mucoid brown colonies of *Cryptococcus* spp

### III. Results

Out of the total 67 clinical samples taken, 16 (23.88%) were CSF, 15 (22.39%) were blood, 17 (25.37%) urine, 16 (23.88%) sputum and 3 (4.48%) others (i.e. aspirate of skin, lymphnode & scalp). Out of 48 patients enrolled in the study, *Cryptococcus* spp were isolated from 16(33.33%). Among these 16 cryptococcosis patients, majority of them presented with cryptococcal meningitis 13 (81.25%), while 1 (6.25%) patient each were presented with cryptococcal lymphadenitis, disseminated cutaneous cryptococcosis and osseous cryptococcosis respectively. The ratio of male to female cryptococcosis patients was 4.3:1 (TABLE 1).



**Figure 3:** Swelling over the right side of scalp in osseous cryptococcosis patient.

**Table 1:** Distribution of cryptococcosis among culture positive patients

Cryptococcosis	Male	Female	Total	Percentage
Cryptococcal Meningitis	11	2	13	81.25%
Cryptococcal Lymphadenitis	1	0	1	6.25%
Disseminated cutaneous Cryptococcosis	0	1	1	6.25%
Osseous cryptococcosis	1	0	1	6.25%
Total	13 (71.43%)	3 (28.57%)	16	100%

Among 16 cryptococcosis patients, 14 (87.5%) were HIV positive. And among these HIV positive patients, route of transmission through sexual was 9 (64.29%) and 5 (35.71%) was through intravenous drug injection. Out of 14 HIV positive patients, 11 (78.57%) were on ART (Table 2)

**Table 2:** HIV prevalence, route of infection and ART status among the culture positive patients in relation to age

Age group	No. of patients	HIV positive	Route of infection		On ART
			STI	IVDU	
1-20	1	0	0	0	0
21-40	11	11	6	5	9
41-60	1	1	1	0	0
61-80	3	2	2	0	2
Total	16	14(87.5%)	9 (64.29%)	5 (35.71%)	11(78.57%)

\*STI-Sexually Transmitted Infection; IVDU-Intravenous Drug Use.

#### IV. Discussion

*Cryptococcus neoformans* has become one of the most prevalent causes of fungal disease affecting more than one million people and causing at least 500,000 deaths annually worldwide due to HIV-associated cryptococcosis [11]. It is suggested that cryptococcosis one of the AIDS defining infections considered as "sleeping disease" became an "awakening giant" within a couple of years [12]. Manipur, located in the north eastern corner of India is one of the high prevalent states of HIV infection [10].

In this present study, the prevalence of cryptococcosis was observed to be high (TABLE 1). This finding is related to the study taken by Uma B [13] which reported increasing trend of cryptococcosis with the increase in the prevalence of HIV/AIDS over the years from 20% to 49%. This high prevalence of cryptococcosis may reflect an enhanced clinical awareness, improved diagnostic capability and also increasing prevalence of HIV/AIDS. Cryptococcal meningitis was the most common presentation (81.25%) followed by cryptococcal lymphadenitis, disseminated cutaneous cryptococcosis and osseous cryptococcosis with 6.25% each respectively. The high prevalence of cryptococcal meningitis patients in this study concurs with the findings reported in some studies in the HIV positive patients [14,15]. And also the prevalence of male higher than the female in this present study was comparable to the reports in other studies [16,17,18]. There is hypothesis that differential interaction between *Cryptococcus* and macrophages within different gender environments and hormone contribute to the increased prevalence of cryptococcosis in males [19].

The percentage of age distribution among cryptococcosis patients was maximum in the age group of 21-40 yrs (68.75%) which is statistically significant (p=0.02) in this study. The maximum distribution of the patients in age group 20-40 was also observed by Lakshmi V et al. [17] with a mean of 31 years. The youngest patient among the study population was a 6 years old boy who presented with cryptococcal lymphadenitis and was HIV negative. A case of disseminated cutaneous cryptococcosis with pulmonary tuberculosis was found in

a HIV positive ART naïve female patient where *Cryptococcus* was isolated from skin aspirates, blood, bone marrow and urine. In this current study, an immunocompetent male patient presented with osseous cryptococcosis on the right scalp. All of them responded well to antifungal therapy. This present study showed that the prevalence of cryptococcosis in HIV positive patients were high (87.5%). The high prevalence of HIV positive in relation to cryptococcosis patients was comparable with the finding of Babbygtonn KDS et al. [16] and the predominance of cryptococcosis in the third and fourth decades of life coincides with the age range where HIV is most prevalent. Among these HIV positive patients, transmission through sexual contact was higher than through intravenous drug injection. Linda M [20] suggested that sexual transmission of HIV is by far the most common mode of transmission globally which is similar with the finding of our study. Out of these 14 HIV positive patients, 11 (78.57%) were on ART (Table 2). The reason for high prevalence of HIV positive cryptococcosis on ART have not been studied here but maybe due to non compliance of the patients or non availability of ART. Manipur presently reporting as high prevalence HIV positive state in India, coupled with its subtropical climate with rich flora and fauna may also be the reason for high prevalence of cryptococcosis. Thus, further study on the impact of ART and exploration of the effect of location and environment on the risk of cryptococcal infection may bring more insight to the current problem.

## V. Conclusion

This study shows that cryptococcosis is highly prevalent especially among HIV positive patients and common presentation being cryptococcal meningitis. In general, people should be made aware of the disease and physicians should have high clinical suspicion especially to HIV positive patients to initiate prompt investigation and treatment for better outcome.

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