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**PREVALENCE OF ASPERGILLOSIS IN PATIENTS OF CHRONIC LUNG DISEASES
IN A TERTIARY CARE HOSPITAL OF NORTH EAST REGION OF INDIA**

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Abstract

Background- Aspergillosis is a systemic infection found in immunocompromised as well as immunocompetent individuals. This is a primary pulmonary infection with involvement of other body sites like paranasal sinuses and cutaneous tissues. This study was conducted to find out the prevalence of Aspergillus infection in patients of chronic lung disease in different age and gender groups.

Materials and Methods- This two year observational study was carried out in the Department of Microbiology and samples were collected from the Department of chest medicine at Agartala Government Medical College, and Panth hospital, Agartala. Sputum samples from 226 patients of chronic lung diseases of different age and gender groups with clinical findings of fungal infections were processed and investigated by direct microscopy and culture.

Results- Aspergillus species were isolated in 38(16.81%) cases of chronic lung diseases. Aspergillus fumigatus was the predominant isolate in 30(78.94%) cases.

Conclusion- The present study was undertaken to describe prevailing spectrum of pulmonary aspergillosis in patients of chronic lung diseases.

Key words- Aspergillosis, prevalence, chronic lung diseases.

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Introduction:

Aspergillus species may cause various type of pulmonary diseases based on the immune status of the individual. Different aspergillus species have been isolated from soil, indoor environment of hospitals, water tanks etc. Aspergillus conidia enter in the body commonly via respiratory tract by inhalation of water aerosols contaminated with aspergillus conidia to lung. Hyphal transformation with vascular invasion and disseminations of infection are potential sequelae¹. With this

understanding, aspergillosis have predilection for specific immune defects and chronic lung diseases. The primary route of infection is through inhalation of the fungal spore. From lung it causes systemic infection after dissemination².

Tuberculosis of lung is predisposing factor for colonisation of fungal spore in lung as documented by some studies³. More over antifungal treatments is given against aspergillus in chronic lung diseases are variable and controversial^{4,5}. Other chronic lung diseases like lung abscess,

asthma, bronchitis, pleural effusion, malignancy are also associated risk factors⁶. Prevalence of *Aspergillus* species in sputum shows greater frequency in tuberculosis patients as compare to malignancy and chronic lung diseases⁷. The diagnosis of *Aspergillus* infection is difficult. The signs and symptoms in most of the cases of aspergillosis are non-specific, and radiological findings are of little diagnostic value. Diagnosis of aspergillosis requires the repeated isolation and identification of the fungus. Routine blood cultures are not much helpful as they are insensitive for diagnosis of aspergillosis⁸. Pulmonary aspergillosis is common in immunocompromised hosts in whom immune response is less or may be absent. Systemic antibody response in immunocompromised patient and its detection is likely to be unreliable indicators of infection⁹. The present study was undertaken to find out the correlation between direct microscopic findings in 10% KOH mount with culture of sputum for diagnosis of *Aspergillus* infection, and to study the prevalence of various species of *Aspergillus* and predisposing factors for *Aspergillus* infection in chronic lung diseases.

Materials & Method:

This observational study was carried out in the Microbiology Department of G.B.Panth Hospital, and Agartala Government Medical college, Agartala. 226 sputum samples were collected from the department of during the period of 2 years (January 2010 to January 2012). Sputum samples were collected from patients of chronic lung diseases with the suspicion of secondary aspergillosis on the basis of clinical and radiological findings in the present study. Sputum samples of two consecutive days were collected from each patient in a wide mouth autoclaved sterile glass bottle and examined by microscopic examination of 10% KOH mount and cultured on Sabouraud Dextrose Agar (SDA) medium with antibiotic chlormphenicol (.05 mg/ml) and incubated at 250 C and 370 C respectively. Primary identification was done on the basis of septate hyphae and dichotomous branching in KOH mount. The culture was examined daily during first week and twice a week for further four weeks. Only those samples were taken in account in which isolates of

Aspergillus were positive in two consecutive cultures. The fungus which grew in the first culture but subsequently did not grow in the second culture was discarded and not documented. Tease mount preparation (TMP) of fungal growth was made in lactophenol cotton blue for identification of morphology of sporing fungus from cultural fungal growth. Microslide cultures were also performed for confirmation of fungal species following standard method. Statistical analysis was done by applying chi square test and using SPSS software.

Results:

Out of 226 samples 45(19.90%) samples were positive under 10% KOH mount by direct microscopy, 38(16.81%) patients showed growth of *Aspergillus* species in two consecutive sputum sample cultures. Statistically no significant difference ($p>0.05$) was found between KOH mount and fungal culture for diagnosis of aspergillosis in patients of chronic lung diseases. (Table 1). Out of 38 culture positive cases 23(60.52%) were positive among the 49-58 years age group (Table 3). Males were predominantly affected having the percentage of 73.68% (Table 3). Male and female ratio was 4:1. Majority of patients (86.72%) presented with history of unproductive cough followed by fever (69.02%),dyspnoea (51.32%), hemoptysis (27.8%), weight loss (22.56%), productive cough (15.04%) and chest pain (11.94%) respectively (Figure 1). Majority of the patients with chronic lung diseases were suffering from pulmonary tuberculosis (114) followed by chronic bronchitis (26), asthma (20) lung abscess (20), pleural effusion (18) bronchogenic carcinoma (16) and bronchopneumonia (12) respectively. Out of 226 chronic lung diseases cases, *Aspergillus fumigatus* was the predominant species isolated from the culture (78.94%), followed by *Aspergillus flavus* (15.78%) and *Aspergillus niger* (5.26%). *Aspergillus* cultures were positive in 26 (22.80%) of pulmonary tuberculosis cases, 6(37.5%) of malignancy cases, 4 (15.38%) of lung abscess cases, and 2 (10.0%) of the asthma cases (Table 2).

Discussion:

In our study, out of 226 patients of chronic lung diseases, 38(16.7%) patients showed the growth of *Aspergillus* species in two consecutive

sputum fungal cultures. This study is close to the findings of Sahid et al where the prevalence of aspergillus species among patients of chronic lung diseases by direct microscopy and fungal culture was found to be 14.7%¹⁰.

The mean age of Aspergillosis in this study was found 45.6±15.4 years and male: female ratio was 4:1, which is similar to the results reported by other studies^{10,11}.

Majority of the patients with chronic lung diseases in our study were suffering from pulmonary tuberculosis (114) followed by chronic bronchitis (26), asthma (20), lung abscess (20) pleural effusion (18) malignancy (16) and bronchopneumonia (12) respectively. Other studies have also described similar findings². Aspergillus cultures were positive in 26 (22.80%) of pulmonary tuberculosis cases, 6(37.5%) of malignancy cases, 4 (15.38%) of lung abscess cases, and 2 (10.0%) of the asthma cases in our study.

Susceptibility of Aspergillus appears to be enhanced by certain factors like therapy with antibiotics and chemotherapeutic drugs and steroids, depression of bone marrow activity, carcinoma and leukaemia. Especially antibiotics and steroids appear to be more significant as they may stimulate the growth and virulence of infecting fungus by destruction of competing bacterial flora. Common use of broad spectrum antibiotics in the pulmonary disease for long duration, for example in the pulmonary Tuberculosis, lung abscess and chronic bronchitis may predispose to fungal infections., Pulmonary tuberculosis is most common predisposing factor for colonising and invasive aspergillosis of lung^{10,12}.

In the present study, Aspergillus fumigatus was the predominant species isolated from the culture (78.94%), followed by Aspergillus flavus

(15.78%) and Aspergillus niger (5.26%). Aspergillus fumigatus is dominant species among other Aspergillus species responsible for pulmonary diseases. Aspergillus genus comprises approximately 180 species out of which 34 species are associated with human diseases. In a study by Morgan et al, the prevalence of Aspergillus fumigatus was documented as 90% in patients of chronic lung diseases¹³. In another study by Morgan et al the prevalence of aspergillus species associated with chronic lung infections were Aspergillus fumigatus (56%), Aspergillus flavus (18.7%) Aspergillus niger (8%) Aspergillus terreus (16%)¹⁴. In the study of Chawla et al Aaspergillus fumigates was documented as 40.9% in patients of chronic pulmonary diseases¹⁵. The findings of our study are similar to above mentioned studies where Aspergillus fumigatus is the dominant pathogen in chronic lung diseases. In our study, significant predilection was observed for pulmonary aspergillosis in patients suffering from Bronchogenic carcinoma. Our study correlates with the study of Biswas et al who also reported significant relation between colonisation of Aspergillus sp. in patients suffering from Bronchogenic carcinoma¹⁶.

Conclusion: It is concluded from the present study of North East region that prevalence of aspergillosis in patients of chronic lung disease is high as compare to other study of India. Therefore outcome of chronic lung disease is poor. So we suggest that for every patients of chronic lung diseases, 10% KOH mount and fungal culture of sputum is utmost needed to establish the correct diagnosis of aspergillosis so appropriate antifungal therapy can be advocated.

Table-1: Sputum microscopy and culture positivity for Aspergillus species among patients with chronic lung diseases (p ≥0.05)

SAMPLES	10% KOH	%	CULTURE	%
Positive	45	19.90	38	16.80
Negative	181	80.10	188	83.20
Total	226	100.00	226	100.00

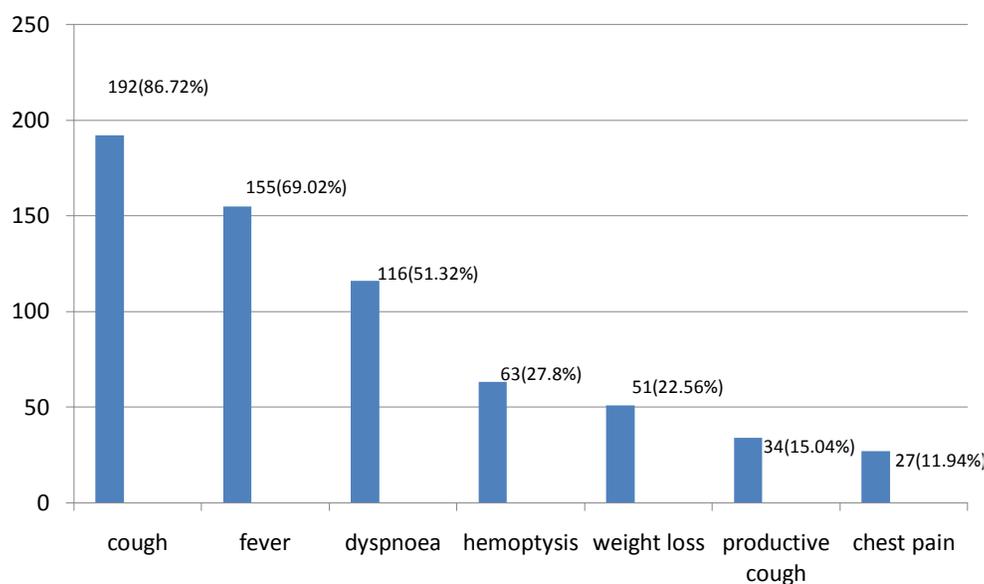
Table-2: Prevalence of different Aspergillus species in various chronic lung diseases

Pulmonary diseases	No. of patients	Aspergillus species isolated from sputum samples			
		A. Fumigates (%)	A. niger(%)	A. flavus(%)	Total
Pulmonary tuberculosis	114	20	4	2	26(26.80%)
Chronic bronchitis	26	-	-	-	-
Asthma	20	2	-	-	2(10.00%)
Lung Abscess	20	2	2	-	4(15.38%)
Malignancy	16	6	-	-	6(37.5%)
Pleural effusion	18	-	-	-	-
Bronchopneumonia	12	-	-	-	-
TOTAL	226	30(78.94%)	6(15.78%)	2(5.26%)	38

Table 3:- The sex and age distribution among total culture positive cases of Aspergillosis

Age groups (years)	Male	Female	Total
18-28	-	1(2.63%)	1
29-38	3(7.89%)	1(2.63%)	4
39-48	5(13.12%)	2(5.26%)	7
49-58	18(47.36%)	5(13.12%)	23
59-68	2(5.26%)	-	2
≥69	-	1(2.63%)	1
Total	28	10	38

Figure 1 : Presenting features of Chronic Lung Diseases patients (n=226)



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