



ROLE OF *ASPERGILLUS* SPP IN EXACERBATION SEVERITY IN CHRONIC OBSTRUCTIVE PULMONARY DISEASE

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ABSTRACT

Introduction: Patients of Chronic Obstructive Pulmonary Disease COPD experience exacerbation of their condition and becoming one of main prospect for developing into Invasive Pulmonary Aspergillosis(IPA). **Aims and Objectives:** To isolate *Aspergillus* genus from COPD Patients suffering from Lower Respiratory Tract Infection and determine its role of exacerbation in them. **Material and Method:** Respiratory samples and whole blood were collected from COPD patients and respiratory samples were cultured on SDA agar for isolation of *Aspergillus* species, which were confirmed by standard mycological methods and presence of specific antibodies of *Aspergillus* screened from whole blood. **Results:** Out of 150 LRTI patients, 45(30%) patients were declared as COPD in which 25(55.5%) were hospitalized and 20(44.4%) were out patients. *Aspergillus* spp were isolated in 23(51.1%) patients only, most of them were hospitalized with exacerbation. **Conclusion:** Colonization of *Aspergillus* increase the severity of exacerbation in COPD patients and worsen the condition if not treated early.

KEYWORDS : Chronic Obstructive Pulmonary disease (COPD), *Aspergillus fumigatus*, Invasive Pulmonary Aspergillosis (IPA), Forced Expiratory Volume(FEV), European Organization for research and treatment for Cancer and Mycoses study Group (EORTC/MSG), Bronchoalveolar Lavage (BAL), Endotracheal Aspirate (ETA)

INTRODUCTION

Chronic obstructive pulmonary disease (COPD) has been identified as risk factor for invasive Aspergillosis, in the recent years, which occurs due to smoking tobacco extended exposure to pollution caused by burning of wood and other biomass fuels, recently it has been estimated to rank by 2020 that COPD is the fourth cause of morbidity and mortality, COPD disease characterized by an airflow restriction and inflammation of the lower airways. Patients with COPD can observe "exacerbation" of their condition. An exacerbation is an event defined subjective descriptions or symptoms, namely dyspnoea, cough, sputum production that aggravate sufficiently to warrant a change in medical management.²

Recent reports have documented expansion in patients with COPD which constitute a less immunodeficient group that are becoming susceptible to invasive aspergillosis specially on steroid therapy, along with patients receiving immunosuppressive therapy although it is preventable and curable usually progressive and irreversible but on the other hand majority of death from COPD is occur in less developing countries.² Bacteria and viruses are the considerable cause of COPD exacerbation whereas the role of fungi is less well understood¹.

Most pulmonary infection caused by member of genus *Aspergillus*. Aspergilli are ubiquitous soil dwelling fungi that have capacity to cause a range of pulmonary conditions including Invasive Aspergillosis, Chronic Pulmonary Aspergillosis including Aspergilloma³, Hypersensitivity pneumonitis and Allergic bronchopulmonary aspergillosis. In the upper respiratory tract, *Aspergillus* can usually colonize where the development of the infection requires the penetration of the spores in the lower airways, which associated with germination and tissue invasion¹.

Colonization and its potential role in Exacerbation of COPD are imperfectly understood, therefore the present study was planned to determine *Aspergillus* colonization in COPD patients is an exacerbation factor.

Place of study

This cross-sectional study was conducted in Department of microbiology of Index Medical college and research centre and Hospital, Indore, MP.

Study Population

This study was carried out at Index Medical College Indore from May 2018 to June 2020. Total 150 patients were visited Pulmonary Department, showing symptoms of lower respiratory tract infection

were selected for the study. Patients with active Tuberculosis, HIV reactive, immunocompromised patients and those with malignancies were excluded from the study. The informed consent was taken from all the study participants.

The study was performed on 45 patients 42 males and 3 females all age above 18- 80 years diagnosed with COPD, as define by Global Initiative for Chronic Obstructive lung Disease guidelines.

METHODOLOGY

The total 45 respiratory samples were collected for the study, which were sputum, BAL(broncho alveolar lavage) by fiberoptic bronchoscopy, ETA (Endo Tracheal Aspirate) in intubated patients. Whole blood were also taken from patients for Antibody detection.

Microscopic examination of all samples were done with 10% KOH mount for identify fungal elements. Respiratory tract specimens were homogenized by adding N-acetyl L- Cystine in M/50 Trisodium citrate and diluting double the amount with phosphate buffer, then centrifuged and only remaining sediment were cultured on Sabouraud, S Dextrose Agar (SDA) with chloramphenicol slants.

All SDA tubes were incubated at 30°C in dark for around 3-4 days. *Aspergillus* isolate were identified according to conventional technique based on growth and standard morphologically characteristics following standard mycological procedure. Serum was separated from whole blood samples and screened for specific antibody of *aspergillus* by using readymade ELISA kits (GENESIS DIAGNOSTICS). Statistical analysis was done with Chi square test USING SPSS software.

RESULTS

Of the total 150 patients suffering from LRTI, 45(30%) were clinically diagnosed with COPD in which 42(93.3%) were males and 3(6.6%) were females, all the patients were above than 18 years and the mean age was 57.7 year. Out of 45 patients 21(46.6%) were having dyspnea, 40(88.8%) were having cough in these 30(66.6%) patients were having more than 12 years of smoking history. Rhonchi and Chest pain complained by 8(17.7%) and 6(13.3%) patients respectively. 2(4.4%) of the patients were suffering from right side pleuritic pain. 8(17.7%) of the patients were going through weight loss. History of tuberculosis(TB) were seen in 8(17.7%) patients and 12(26.6%) were having corticosteroid therapy (Shown in **graph 1**)

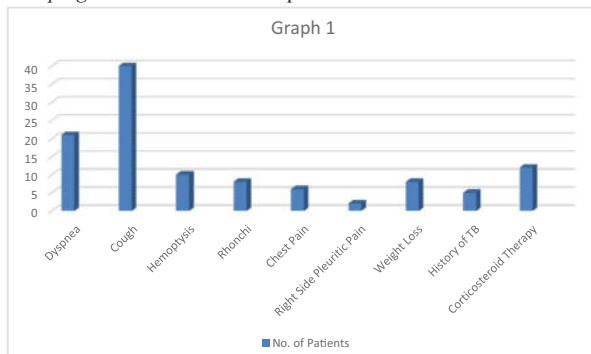
Duration of smoking were reported in 30 patients while mean duration of 59 pack per year (range 20-120 pack per year) 5(16.6%) patients

have severe COPD, their spirometry test have also done. The mean FEV1 (Forced expiratory volume in 1 second) was 1.18 L (range 0.75L to 2.15 L). 4(13.3) patients were documented recent acute exacerbation of COPD. 12(40%) were on chronic systemic corticosteroid therapy. The mean corticosteroid dose 12 mg per day (range 15 to 65)mg per day.

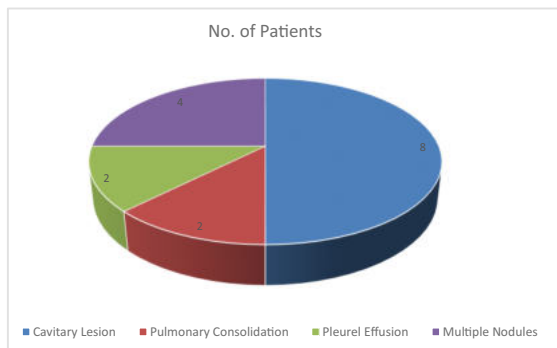
The radiological evaluation was based on chest radiographs. The most commonly reported radiological abnormality were cavitory lesion, infiltrate or consolidation were reported 2 (4.4%)patients, pleural effusion and multiple nodules were found in 2 (2.2%)and 4 (8.8%)patients respectively (Shown in **graph 2**).

Direct examination and culture of respiratory specimen yielded positive outcome for 23(51%) of the samples colonization with *Aspergillus* species. Of the 23 *Aspergillus* isolate, 15 (65%)were *Aspergillus fumigatus*, 6(26%)were *Aspergillus flavus* and 2(8%) were *Aspergillus niger*. *Candida* species were found in 8 (17%)samples, of which 6 (75%)were *candida albican* and 2(25%) were *candida tropicalis*. Among the species *Aspergillus fumigates* is most isolated specie in study (shown in **graph 3**).

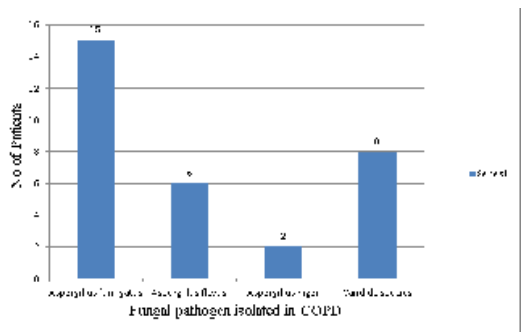
Aspergillus fumigates IgG detection also screened from patient serum sample, which were also culture positive for *Aspergillus fumigates*. Out of 45, 11 (24%)patients have raised IgG antibody for *Aspergillus fumigatus*. Stable patients who were visited OPD only and found to be negative for exacerbations but found cultured positive for *Aspergillus*, their condition degraded with the time. 12 (26%)unstable patients admitted in ICU had exacerbation of COPD, yielded 10(83%) *Aspergillus* species. The severity of exacerbations could be the marker of *Aspergillus* infection in COPD patients.



Graph 1: Clinical Representation of COPD Patients.



Graph 2: Distribution of Radiological finding of COPD patients.



Graph 3: Species isolation among COPD patients.

DISCUSSION

COPD is chronic inflammatory disease of pulmonary air way which is most wide spread non communicable disease of the lungs. It is both preventable and treatable but generally progressive and irreversible with majority of death from COPD occurring as well as in less developed countries¹⁰.

The development of COPD is has numerous reasons. One factor can be the genetic and environment factor can also be there. Hammond et al states that causation elements in the onset of reducing lung functions and progression of COPD Biomass fuels and mining .

This disease reversibly and progressively limits airflow which results in defective immune response of lungs to various particles. The word “Exacerbation” is always connected with COPD It is a sign and one of the symptoms in COPD patients when the lungs respond to infection or irritating substance by developing inflammation that made the airways tapered from muscles- tightness, swelling and mucus. the sign and symptoms of Exacerbation is caused by the changes in the airways caused which will help you to get treatment before time and shorten the length of Exacerbation. The length of Exacerbation also indicated on the severity of infection or inflammation in lungs. If Exacerbation length is short, it will prevent and cure before becoming severe.²⁶

Using Inhalers, steroids or antibiotics Some exacerbation can be treated at home , patients will need to be hospitalized if symptoms become serious . Treatment of an exacerbation is primarily sway the swelling and constriction in your airways and most important to find out the causative agent of it²⁵.

In the present study almost 25(55%) patients reported with exacerbation, having cough dyspnea history from longer time with wheezing sound with it. Of the 25 patients only 16 (64%) patients sputum yielded *Aspergillus* as one of their exacerbation reason in lungs,5 (31%) of them also had past history of Tuberculosis.

Dakshina et al, also reported the 55% of hospitalized COPD patients had exacerbation and worsened clinical conditions were observed.

It was found that 25(55%) of hospitalized patients 6(24%) were having cavitory lesions. 12 (48%)out of them on systemic corticosteroids therapy and 2(8%) of them were on inhaled corticosteroids.

Muquim et.al. and Dakshina et al in similar study also observed that prolong use of corticosteroids before hospitalization in COPD cases associated with growth of invasive pulmonary *Aspergillosis*. Tong. et. al also reported that high dose corticosteroids use were a risk of *Aspergillus* colonization or positive *Aspergillus* culture.^{20,28}

In the present study males were more affected than females. 42(93%) were males and only 3 (6%)were females among COPD patients with LRT.I The higher predilection of LRTI are males as compared to females may be due to their more outer exposure to outside environment .

In our study 30(66%) patients were smokers, 5(16%) of them are chain smoker(5 pack/day)6(20%) were farmers 2(6%) of them are labourer in some chemical factory, all of them having some symptoms of LRTI.

Similar study by Vijay et. al which focus on prevalence of LRTI in patients presenting productive cough, found males to be at high risk of tobacco and alcohol consumption that results lower Mucocillary defenses airway collapse and respiratory fatigue high risk¹⁹.

Tong et. al state that cigarette smoking and continuous Airway inflammation could alter the structure and functioning of lungs and injure a profound effect on the host defence against invading pathogens and particulates, while impairing the airway epithelium and these COPD patients are most susceptible to *Aspergillus* colonization. Luigi et. al in Respiratory research 2006 clears that smoker patients for the longer the higher the likelihood that excess evasion will be more severe^{23,26}.

Among the species *Aspergillus fumigatus* and *Aspergillus flavus* were the most isolated common pathogen. In present study *Aspergillus fumigates* was the major isolated among the patients with possible IPA.

Which is similar to study by Dakshina et al and unlike other studies which observe *Aspergillus flavus* as predominant species.

In our study we found that *Aspergillus* colonization in COPD patients make them susceptible for the progression of exacerbation and the development of IPA. A similar study by Dakshina et al. also reported the same. In contrast a study by Guinea et al. found COPD as a predisposing state for the development of IPA in these hospitalized patients. The main reason we think, for this exceedingly high mortality in COPD patients is related to delay in the diagnosis of IPA, because it is not routinely applied in this population. In literatures it has been displayed that early diagnosis of IPA is associated with better outcome, relieve the IPA in COPD patients would probable improve by increased awareness of this serious infection in this patient population, most those with severe COPD and on chronic corticosteroid therapy.

No operative surgery or biopsies of any part including lungs not done in our research we found our patients to be colonized, since none of our patients met the norm for "proven" or "possible" invasive fungal disease based on the particulars of invasive fungal disease propounded by the EORTC/MSG and classifying the cases IPA only as predictable and believable IPA, which is in contrast to a study by Bulba et al. to characterize the LRTI patients into proven probable and possible, although the clinical conditions and imaging finding of IPA and COPD patients are no indifferent, both condition have similar feature and predisposing factor²⁷. It has been also postulated that *Aspergillus* isolated from any of patients, hospitalized or OPD patient, critically ill (even when immunocompetent), elder patients should not be rejected as colonization, especially in old age patients it can be underline disease. Isolation is usually explained as colonization, but confirmation can only be done by histopathological evidences, that is not possible in these patients⁴.

CONCLUSION

The study concludes that there is a direct and perceivable relation between the exposure, colonization and isolation of *Aspergillus* from COPD patients and *Aspergillus* colonization in unstable chronic smoker COPD patients being a reason of exacerbation in them. The severity of exacerbation also reveals condition of colonization in COPD patients, Prolong treatment with chronic corticosteroids therapy is one of the evolve reason. Early non invasive diagnosis becomes very challenging and difficult to initiate timely to save lives because of invasive nature of fungus and mortality associated with it²⁵. Studies also evaluating the outcome of elders COPD patients with *Aspergillus* species are increasing and required to be isolated early along with adequate antifungal therapy.

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