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Prevalence of Different Species of Candida in Sputum of Pulmonary Tuberculosis

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A B S T R A C T

Pulmonary tuberculosis is one among the most common disease in developing countries. *Candida species* infection has always seemed to be associated with secondary infections in Tuberculosis. *Candida species* have been one of the co-infection challenges facing the patients suffering from pulmonary tuberculosis. Hence this study was undertaken to find out the prevalence of *Candida species* in cases of pulmonary tuberculosis. This prospective study was carried out in Karnataka institute of medical sciences, Hubballi over a period of one year, from January 2012 to December 2012. 100 patients were included whose sputum were tested positive in Designated Microscopy Centre (DMC) as per the RNTCP guidelines. The positive sputum samples were collected and further processed for microscopy (gram stain for sputum quality), fungal culture on SDA with and without cycloheximide. [*Candida* co-infection was confirmed using the (Kahanpaa, 1972) criteria] Out of the total 100 patients with pulmonary tuberculosis**,** whose sputum were tested positive in Designated Microscopy Centre (DMC) as per the RNTCP guidelines, Candida co-infection was observed in 32 (32%) of patients. There was a significant male preponderance for Candida co-infection. *Candida albicans* and *Candida tropicalis* are the most commonly isolated species 11 (34.4%), followed by *Candida parapsilosis* 7 (21.9%), *Candida dubliniensis* 2 (6.2%) and *Candida glabrata* 1 (3.1%). The coexistence of fungal with tuberculosis adds complication to patient's condition by adding more damaging and fatal dimensions to it. So identification of *Candida* species is essential as they differ in their virulence and sensitivity to antifungal drugs.

Introduction

Tuberculosis (TB) causes significant morbidity and mortality throughout the world, particularly in developing countries in Asia and Africa. At present about one third of the human population is infected with *Mycobacterium tuberculosis* and every year two million persons die because of it (Rathod *et al.,*) The World Health Organisation (WHO) TB statistics for India for 2015 give an estimated prevalence is given as 2.5 million. It is estimated that about 40% of the Indian population is infected with TB Bacteria, the vast majority of whom have latent TB rather than TB disease. (www.tbfacts.org)

Candida species are emerging as a potentially pathogenic fungus in patients with broncho-

pulmonary diseases. The synergistic growth promoting association of *Candida* and *Mycobacterium tuberculosis* has raised increased concern for studying the various Candida species and its significance in pulmonary tuberculosis patients during current years (Kali *et al.*, 2013).

Hence, this study was undertaken with the objective of finding the prevalence of co-infection caused by different Candida species in patients with pulmonary tuberculosis.

Materials and Methods

This prospective study was carried out in Karnataka institute of medical sciences, Hubballi over a period of one year, from January 2012 to December 2012. 100 patients were included whose sputum were tested positive in Designated Microscopy Centre (DMC) as per the RNTCP guidelines.

The positive sputum samples were collected and further processed for microscopy (gram stain for sputum quality), fungal culture on SDA with and without cycloheximide. (*Candida* co-infection was confirmed using the (Kahanpaa, 1972) criteria)

The growth of cream pasty opaque colonies >30 colonies on SDA slant is considered significant and confirmed by gram stain. Gram stain showing gram positive budding yeast cells with pseudohyphae. The colonies further subjected to germ tube test morphology on corn meal agar, sugar fermentation tests for speciation.

Results and Discussion

Out of the total 100 patients with pulmonary tuberculosis**,** whose sputum were tested positive in Designated Microscopy Centre (DMC) as per the RNTCP guidelines, Candida co-infection was observed in 32 (32%) of patients. There was a significant male preponderance for Candida co-infection (Table 1). *Candida albicans* and *Candida tropicalis* were the most common isolate observed in 34.4% of patients with Candida co-infection followed by *Candida parapsilosis* (21.9%) (Table 2).

Tuberculosis is well recognised for its wide range of clinical spectrum, chronicity and sequlae (Ndukwu *et al.*, 2016). Pulmonary tuberculosis is essentially a chronic destructive disease of the lungs. Caseation, necrosis and fibrosis lead to the formation of cavities with bronchiectatic dilations. These cavities form an ideal culture plate for the tubercle Bacilli and many other organisms including the fungi by providing plenty of oxygen along with necrotic tissue material.

Moreover, the prolonged antitubercular therapy in tuberculosis, which may last for well over 2 years with or without corticosteroids by itself, becomes a potent predisposing factor for the onset of superinfection by the fungal organisms.

Therefore, it is not surprising to come across frequent references in the literature regarding the association of mycological superinfection with active pulmonary tuberculosis. (Astekar *et al.,* 2016)

Considering the huge population of TB patients, a large number of individuals are at the risk of fungal infections. Coexistence of *Candida* and tubercle bacilli is known since a long time (Rathod *et al.,*). Though several authors have documented *Candida* species as the most common fungal agent isolated from sputum of pulmonary tuberculosis patients, its significance has always been a matter of controversy due to the fact that up to 32.5% healthy people carry *Candida* in their throat. This can contaminate the sputum sample during collection. (Santiwongkarn *et al.,* 2012)

Table.1 Gender wise distribution of the study population

Gender	Number	Percentage (%)
Male	75	75
Female	25	25
Total	100	100

Table.2 Distribution of different species of *Candida* in patients with pulmonary tuberculosis

Candida species	Number	Percentage (%)
Candida albicans	11	34.4
Candida tropicalis	11	34.4
Candida parapsilosis	7	21.9
Candida dublinensis	2	6.2
Candida glabrata	1	3.1
Total	32	100.0

To eliminate contamination, various approaches have been used like bronchoscopy sample (El-Ebiary *et al.,* 1997). Jain *et al.,* (1982) have compared and subtracted the growth of *Candida spp.* on plain SDA from mouth rinsed water as a control to that of sputum sample, criteria suggested by Kahanpaa (1972). According to this criteria three or more repeated isolations of *Candida* more than 30 colonies on SDA with pseudomycelial forms in sputum microscopy is more suggestive of infection than colonisation. In this study we have also followed this criteria to detect *Candida* infection.

The prevalence of *Candida* co-infection of lung ranges between 15-32% in different studies (Jain *et al.,* 1982). *C. albicans* has been reported to be the most predominant isolate from sputum of tuberculosis patients followed by *C. tropicalis* (Latha *et al.,* 2011). In our study, out of the total 100 patients with pulmonary tuberculosis, whose sputum were tested positive in Designated Microscopy Centre (DMC) as per the RNTCP guidelines, Candida co-infection was observed in 32 (32%) of patients which is similar to study by Kali *et al.,* (2013) *Candida albicans* and *Candida tropicalis* are the most commonly isolated species 11 (34.4%), followed by *Candida parapsilosis* 7 (21.9%), *Candida dubliniensis* 2 (6.2%) and *Candida glabrata* 1 (3.1%) in the sputum samples collected from patients diagnosed with pulmonary tuberculosis.

The results obtained from this study are similar to results obtained by Kali *et al.,* (2013) where 50% of *C. albicans*, 20% of *C. tropicalis*, 20% of *C. glabrata*, 6.7% of *C. parapsilosis were isolated* (Pandalai and Kurup, 1962), isolated 6 (8.7%) of *Candida parapsilosis* out of 69 cases. Lata *et al.,* (2011) documented *Candida tropicalis* (19.95%), *Candida glabrata* (16.54%), *Candida parapsilopsis* (13.14%). Jain *et al.,* (1982) reported *C. tropicalis* (9.1%). Baradkar *et al.,* (2009) detected *C. tropicalis* 3.25%, *Candida parapsilosis* 3.25%.

These variations in percentages are mainly attributed to differences in local prevalence of different species due to different environmental conditions, as well as to the various detection methods employed.

Among the LRTIs, tuberculosis has long been a major public health challenge in developing countries. Pulmonary mycoses relatively by themselves are not grossly damaging; however, when superimposed in conditions such as tuberculosis, their impact on the morbidity and mortality pattern renders them vitally important.

Fungal infections of lungs are important infective processes. Candida infection has been the most important secondary infection.

Diseases like opportunistic infections if it is diagnosed early can be treated and thus can prevent progression to fibrotic stage. We found increased prevalence of non-albicans *Candida spp* in our study in pulmonary tuberculosis patients in the hospital. Taking this account the present study tries to assess some of aspects of pulmonary mycosis.

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