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SARS CoV-2 VIRUS Vs OPPORTUNISTIC MYCOSES

Microbiology

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ABSTRACT

Purpose: SARS CoV 2 virus a novel coronavirus, which is a single stranded positive sense RNA virus. It causes severe pneumonia in susceptible individuals. In individuals with predisposing factors, like diabetes, neutropenia, corticosteroids therapy, and etc., the virus causes opportunistic fungal infections. This study aims the correlation between COVID 19 disease and fungal infections.

Methodology: The present study includes 202 specimens collected from patients with symptoms suspected of fungal infections. The collected specimens were subjected to direct microscopy and fungal culture.

Results: Nasal tissue scrapings were the most common specimen collected followed by sputum. Among the 202 specimens collected, 106 were positive for direct microscopy and 81 were positive for fungal culture.

Conclusion: The fungal infections are more common among the patients with comorbidities, and appropriate guidelines should be followed in the management of such infections in order to prevent the morbidity and the mortality.

KEYWORDS

COVID 19, Opportunistic infections, Mucormycosis, Invasive Candidiasis, Aspergillosis.

INTRODUCTION:

At the end of the year 2019, a viral borne pneumonia like infection which initially occurred in Wuhan province, China and rapidly spread across the world.[1] The WHO declared the outbreak a Public Health Emergency of International Concern on 30.1.2020. Based on genome sequencing technology the International Committee on Taxonomy of Viruses [ICTV] announced "SARS-CoV-2" as the name of the new virus on 11.2.2020. It was declared a pandemic by WHO on 11.3.2020.[2]

As of June 2021, there have been 173609772 confirmed cases of COVID-19, including 3742653 deaths around the world.[2] The coronaviruses are enveloped positive sense single stranded RNA viruses which binds to the human Angiotensin Converting Enzyme-2 receptors present on the humans organs or tissues like heart, lung, kidney, bladder, eyes, etc to gain entry and trigger a wide range of clinical manifestation. The COVID-19 disease from human to human through droplets. The presence of the binding receptors in the different tissues determines the various range of clinical manifestations that range from a self limiting illness with general complaints like fever, cough, and headache to more severe complications including Acute respiratory distress syndrome, cardiac arrest, liver failure, and multi organ failure.[1]

COVID-19 is also associated with a wide range of opportunistic bacterial and fungal infection. The predisposing factors being Type 2 Diabetes mellitus, Neutropenia, low oxygen levels, steroid induced hyperglycemia, ketoacidosis, increased iron levels and decreased phagocytic activity of WBC due to immunosuppression.[3]

Aspergillus and Candida have been reported as the most common fungal pathogen in the patients co-infected with COVID-19. Recently there is an increase in Mucormycosis cases seen in patients co-infected with COVID-19.[4]

Mucormycosis are caused by the fungi which belong to Order Mucorales. They are broad or ribbon shaped of 5 to 15 micron in width, irregularly branched, pauciseptate or aseptate molds that reproduce sexually through the formation of zygospores.[5] Globally the prevalence of Mucormycosis is 0.005 to 1.7 per million population, while the prevalence has increased in India to 0.14 per 1000 population, in a recent estimate in the year 2019 to 2020.[3]

The aim of this study is to study the prevalence of fungal infections in post COVID patients presenting with any symptoms pertaining to any of the fungal infections.

MATERIALS AND METHODS:

A cross sectional study was done at SVRRGGH over a period of two months from May 2020 to June 2020 in the Department of Microbiology, S.V. Medical College, Tirupati, after obtaining permission from the Institutional Ethical Committee. The total study participants were 202. The study included, patients with either RT-PCR confirmed COVID 19 positive or those recovered from COVID 19 infection, with signs and symptoms of invasive fungal infection from various departments. The demographic details, diagnosis of the patients were obtained, recorded and analyzed.

The specimens collected from the patients were sent to the Microbiology laboratory as early as possible. The different types of specimens included, sputum, nasal cavity scrapping, growth from sinuses, corneal scrapping, etc. The tissues samples were collected in a sterile test tube containing normal saline and sent to the laboratory. The specimens were subjected to direct microscopy and culture methods. Microscopy was performed using KOH method, and the specimens were inoculated onto Sabouraud Dextrose Agar (SDA) slopes and incubated in BOD incubator for five days[6]

The positive cultures were identified by their macroscopic and microscopic characteristics. Based on the colony morphology, either Gram stain or Lactophenol Cotton Blue mount is performed. The genus of the organism is identified based on the microscopic appearance of colony morphology. The diagnosis was confirmed by histopathological examination and imaging studies.

RESULTS:

The study included specimens from 202 patients among which 134 (66.33%) were male and the most affected age group were 40 to 49 years (29.2%).

Among the various departments were the specimens were collected, 142 (70.2%) of them were from the department of Otorhinolaryngology followed by Department of Pulmonary medicine [CHART 1].

Most of the specimens were collected from patients with post COVID-19 with suspected mucormycosis (89.6%). The common specimens received were nasal cavity scrapings (68.8%) followed by sputum (16.8%) [TABLE 1].

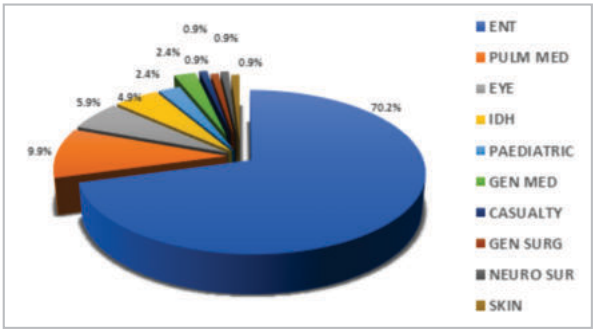

CHART 1: DEPARTMENT WISE DISTRIBUTION OF STUDY SPECIMENS

TABLE 1: DISTRIBUTION OF VARIOUS SPECIMENS INCLUDED IN THE STUDY

TYPE OF SPECIMENS	NO. OF SPECIMENS RECEIVED
Nasal Cavity scrapings	139
Sputum	34
Tissue from sinuses	5
Nasal swabs	4
Corneal scrapings	6
Bronchoalveolar lavage	1
Aspirate from lung abscess	1
Blood	2
Pleural fluid	3
Pus	3
Vitreous fluid	1
Ear swab	1
FNAC from Rt. Lung cavity lesion	1
Skin scrapings	1
TOTAL	**202**

Among the 202 specimens received, KOH mount was performed on 200 specimens and culture was performed on 179 specimens. Among the 200 KOH mount performed, fungal elements were seen in 106 (53%) specimens. Whereas, among the 179 fungal cultures, 81 (45.2%) had growth on the SDA slopes.

Among the KOH mount positive specimens (106), most commonly reported were broad aseptate hyaline hyphae (67.9%), followed by budding yeast like cells (16%) [TABLE 2]. Among the positive fungal culture (81) specimens, more commonly isolated organisms were *Candida* (50.6%) followed by *Rhizopus* (19.7%) [TABLE 3]

TABLE 2: RESULTS OF KOH MOUNT (n=106)

KOH MOUNT RESULTS	POSITIVE	PERCENTAGE
Broad Aseptate Hyaline Hyphae	72	67.9%
Budding Yeast Like Cells	17	16.0%
Narrow Septate Hyaline Hyphae	10	9.4%
Mixed infection	7	6.6%
TOTAL	**106**	

TABLE 3: DISTRIBUTION OF ORGANISMS AMONG POSITIVE SAMPLES (n=81)

ORGANISM ISOLATED	POSITIVE	PERCENTAGE
Candida spp	41	50.6%
Rhizopus spp	16	19.7%
Mucor spp	8	9.8%
Aspergillus spp	8	9.8%
Syncephalastrum spp	5	6.1%
Cunninghamella spp	2	2.4%
Mixed infection	1	1.2%
TOTAL	**81**	

From the total 202 specimens, both KOH and culture positivity were seen in 59 specimens, whereas 47 specimens were positive for only KOH mount and 22 were only culture positive[TABLE 4].

TABLE 4: DISTRIBUTION OF POSITIVE SPECIMENS

KOH MOUNT	FUNGAL CULTURE		
	POSITIVE	NEGATIVE	TOTAL
POSITIVE	59	47	**106**
NEGATIVE	22	74	**96**
TOTAL	**81**	**121**	**202**

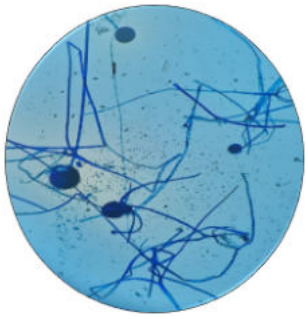

IMAGE 1: LPCB MOUNT DEMONSTRATING RHIZOPUS UNDER LOW POWER MICROSCOPE

DISCUSSION:

SARS CoV -2 virus causes COVID 19 infection which is associated with opportunistic infections as stated by many studies around globe. The present study includes 202 specimens from various departments. Among these 128 (63.4%) were positive for fungal infections. Among the 128 positive specimens, males were more affected 85/128 (66.4%). This correlates with the study by Singh et al[3], where 78.9% of males were affected.

The most common affected organ was nose and sinuses (75%) in the present study, which is similar to the study by Singh et al[3]. The most common sinus affected in the present study is Maxillary sinuses.

In the present study, from the 202 specimens collected, direct microscopy was positive in 52.5% while culture was positive in 40.1%. Patel et al[7], study shows that direct microscopy was positive in 87.3% of specimens while culture was positive in 62.4%.

The most common organism detected in the present study is *Candida* spp. followed by *Rhizopus* spp. Which correlates with study by Chen et al[8].

The limitations of the present study is that, antifungal susceptibility testing was not performed, molecular techniques like PCR was not performed to confirm the diagnosis.

CONCLUSION:

SARS CoV – 2 virus causes a wide range of clinical manifestations from flu like illness to severe pneumonia. The COVID 19 disease is associated with various opportunistic fungal infections among the individuals with predisposing factors. The fungal infections includes Invasive Candidiasis, Mucormycosis, and Aspergillosis. These infections are treated with antifungals like Amphotericin B and azoles like Posaconazole, Isavuconazole etc., which are given for more than four weeks. This results in financial catastrophe for the family and the community as well. In order to prevent such devasting conditions, each institution should prepare a flow chart, which helps the clinicians and the laboratory personnel of how to manage fungal infections in the COVID 19 patients with comorbidities.

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