Assessment of asymptomatic fungal infections in COVID-19 positive and COVID-19 negative pneumonia: A comprehensive epidemiological analysis

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ABSTRACT

Background: The COVID-19 pandemic has underscored the impact of secondary fungal infections on patient outcomes. This single-center study explores asymptomatic fungal infections in COVID-19-positive and COVID-19-negative pneumonia cases, elucidating the intricate relationship between viral respiratory illnesses and fungal co-infections. **Methods:** Conducted at a single center, this cross-sectional study examines the prevalence, risk factors, and clinical implications of asymptomatic fungal infections in patients with COVID-19-positive and COVID-19-negative pneumonia. Various demographic, clinical, and laboratory parameters were analyzed. **Results:** Fungal infections were significantly more prevalent in COVID-19 positive pneumonia cases (60%) compared to COVID-19 negative pneumonia cases (36%), with a notable *P* value of 0.016309. Furthermore, COVID-19-positive patients exhibited distinct clinical characteristics, including increased use of remdesivir (94%), higher rates of invasive mechanical ventilation (36%), and a prolonged hospital stay (14.29 days). **Conclusion:** This study sheds light on the heightened vulnerability to asymptomatic fungal infections in COVID-19-positive pneumonia cases. Understanding these infections prevalence and their associated factors is crucial for comprehensive patient care and may influence treatment strategies, emphasizing the need for targeted interventions in viral respiratory illnesses.

Keywords: COVID-19, fungal infections, hospitalized patients, ICU, immunocompromised, pneumonia

Introduction

The COVID-19 pandemic, precipitated by SARS-CoV-2, presents formidable global health challenges. Emerging evidence underscores the significance of secondary fungal infections in contributing to COVID-19 morbidity and mortality.[1] This single-center study undertakes a comprehensive epidemiological analysis of asymptomatic fungal infections in both COVID-19-positive and COVID-19-negative pneumonia cases, highlighting the intricate interplay between viral respiratory illnesses and fungal co-infections.

Recent investigations have drawn attention to COVID-19 patients susceptibility to opportunistic fungal infections, notably those caused by Candida species and Aspergillus. The immunomodulatory effects of SARS-CoV-2, coupled with the widespread usage of immunosuppressive therapies, create

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a conducive environment for fungal proliferation. Despite often being underestimated, asymptomatic fungal infections significantly impact patient outcomes.[2,3]

Studies by Bartoletti *et al.*[2] and van Arkel *et al.*[4] have further emphasized the association between COVID-19 and invasive pulmonary aspergillosis, shedding light on the clinical implications and management challenges. Additionally, ongoing research by Garcia-Vidal *et al.*[5] highlights the incidence of co-infections and superinfections in hospitalized COVID-19 patients, providing insights into the multifaceted nature of disease progression.

This study, conducted at a single center, delves into the prevalence, risk factors, and clinical implications of asymptomatic fungal infections in both COVID-19-positive and COVID-19-negative pneumonia cases. The objectives include evaluating the prevalence of asymptomatic fungal infections in ICU patients with COVID-19-positive and COVID-19- negative pneumonia, as well as investigating the associated clinical characteristics and risk factors, distinguishing between the two groups.

Material and Methods

Study setting

This cross-sectional study was conducted at SGMH Rewa, encompassing all available Intensive Care Units (ICUs). The study included 50 individuals diagnosed with COVID-19-positive pneumonia and 50 individuals with COVID-19-negative pneumonia. Ethical approval for the study was obtained from the institutional review board.

Data collection

1. Clinical data:
   - Demographic information, including age and gender.
   - Comorbidities and relevant medical history.
   - COVID-19 status (positive/negative) and severity of pneumonia.
   - Use of immunosuppressive therapies and antibiotics.
2. Sample collection:
   - Nasal and oral swabs were collected from each participant.
   - Swabs were processed for direct microscopic examination using potassium hydroxide (KOH) mounts to detect fungal elements.
3. Laboratory analysis:
   - KOH mounts were prepared from nasal and oral swabs.
   - Microscopic examination was performed for the presence of fungal elements, including hyphae, conidia, and yeast cells.
4. Statistical analysis:
   - Descriptive statistics were calculated for demographic and clinical variables.
   - Comparative analysis of fungal infection rates between COVID-19-positive and COVID-19-negative pneumonia groups was conducted using appropriate statistical tests.

Quality control

1. Sample processing:
   - Strict protocols were followed for sample collection to minimize contamination.
   - Laboratory personnel were trained to ensure standardized processing.
2. Microscopic examination:
   - Microscopic examinations were performed by experienced technicians.
   - Regular quality control checks were implemented to ensure accuracy and reliability.

Results

The results of the comparative analysis between patients with COVID-19 positive pneumonia (n = 50) and COVID-19 negative pneumonia (n = 50) are presented in Table 1.

The average age of patients with COVID-19 positive pneumonia was 52.8 years (±14.9), while the average age for those with COVID-19 negative pneumonia was 58.7 years (±15.8).

In COVID-19 positive pneumonia, 29 patients were male (58%) and 21 were female (42%). whereas in the COVID-19 negative pneumonia, there were 30 male (60%) and 20 female (40%).

Figure 1 shows that Fungal infections were significantly more prevalent in patients with COVID-19 positive pneumonia (60%) compared to those with COVID-19 negative pneumonia (36%), with a *P* value of 0.016309. This significant difference highlights a potential association between COVID-19 positivity and an increased risk of fungal infections.

The incidence of diabetes did not differ significantly between the two groups, with rates 42% of patients in COVID-19 positive pneumonia and 30% in the COVID-19 negative pneumonia (*P* = 0.2113). This suggests that the presence of diabetes is not a distinguishing factor between COVID-19 positive and negative pneumonia cases.

Similarly, the prevalence of hypertension showed no significant difference, with rates of 36% in the COVID-19 positive pneumonia and 22% in the COVID-19 negative pneumonia (*P* = 0.122914). This indicates a comparable distribution of hypertension in both groups.

The occurrence of chronic obstructive pulmonary disease (COPD) was also comparable, with rates of 18% in the COVID-19 positive pneumonia and 14% in the COVID-19 negative pneumonia (*P* = 0.585379). This suggests that COPD may not be a distinguishing factor in differentiating the two pneumonia groups.

Although not statistically significant, there was a trend towards significance in corticosteroid use, with rates 90% of patients

| Table 1: Clinical and Demographic characteristic of COVID-19 positive and COVID-19 negative pneumonia			
Characterstics	Covid-19 positive pneumonia (*n*=50)	Covid-19 negative pneumonia (*n*=50)	*P*
Age	52.8+14.9	58.7+15.8	NA
Male/Female	29 (58%)/21 (42%)	30 (60%)/20 (40%)	NA
Fungal infection	30 (60%)	18 (36%)	*P*=0.016
Diabetes	21 (42%)	15 (30%)	*P*=0.211
Hypertension	18 (36%)	11 (22%)	*P*=0.122
COPD	9 (18%)	7 (14%)	*P*=0.585
Corticosteroid	45 (90%)	38 (76%)	*P*=0.062
Remdesivir	47 (94%)	23 (46%)	*P*<0.00001
Tocilizumab	8 (16%)	2 (4%)	*P*=0.045
Invasive Mechanical Ventilation	18 (36%)	8 (16%)	*P*=0.022
Duration of Stay	14.29+8.5	8.9+8.1	*P*=0.002
CT Score	19.94+3.9	17.2+3.2	*P*=0.0002
CRP	52.49+26.8	42.49+24.06	*P*=0.050
D-Dimer	4469.3+2634.3	3321.3+2887.7	*P*=0.040
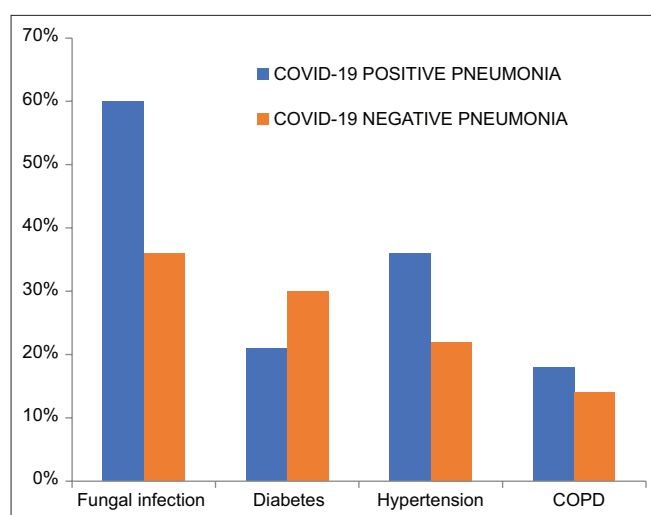
Figure 1: Co morbid medical conditions associated with Pneumonia

in the COVID-19 positive pneumonia group and 76% in the COVID-19 negative pneumonia ($P = 0.062389$).

Highly significant differences were observed, with 94% of COVID-19 positive pneumonia patients receiving remdesivir, compared to 46% in the COVID-19 negative pneumonia group ($P < 0.00001$).

A statistically significant difference was observed in tocilizumab use, with 16% in COVID-19 positive pneumonia and 4% in COVID-19 negative pneumonia ($P = 0.0455$).

Invasive mechanical ventilation was required in 36% of COVID-19 positive pneumonia patients, compared to 16% of those with COVID-19 negative pneumonia, a statistically significant difference ($P = 0.0226$).

The average duration of hospital stay was significantly longer in COVID-19 positive pneumonia cases (14.29 days) compared to COVID-19 negative pneumonia (8.9 days) with a *P* value

of 0.0016. This suggests that COVID-19 positivity may be associated with prolonged hospitalization duration.

Additionally, significant radiological differences were observed, with a CT score of 19.94 ($\pm$3.9) in the COVID-19 positive pneumonia, compared to 17.2 ($\pm$3.2) in the COVID-19 negative pneumonia ($P = 0.0002$). This indicates distinct radiological characteristics between the two pneumonia groups.

The mean CRP level was 52.49 ± 26.8 in COVID-19 positive pneumonia compared to 42.49 ± 24.06 in COVID-19 negative pneumonia ($P = 0.05$). This suggests a potential association between CRP levels and the type of pneumonia, indicating a higher inflammatory response in COVID-19 positive cases.

The D-dimer levels exhibited a statistically significant difference between the two groups. Specifically, the mean D-dimer level was 4469.3 ± 2634.3 in COVID-19 positive pneumonia patients, compared to 3321.3 ± 2887.7 in COVID-19 negative pneumonia patients ($P = 0.0404$). This suggests a notable distinction in the coagulation profiles between the two pneumonia types, with higher D-dimer levels being associated with COVID-19 positive cases.

Discussion

The comparative analysis of COVID-19 positive pneumonia and COVID-19 negative pneumonia patients provides valuable insights into the multifaceted landscape of respiratory infections. These findings highlight several key aspects that warrant nuanced consideration and discussion, aligning with the existing literature on respiratory infections, fungal complications, and COVID-19.

Age and gender distribution

Our results indicate a significant age difference between COVID-19 positive and negative pneumonia patients, aligning with previous studies that underscore age as a crucial factor in COVID-19 severity.[6] However, gender distribution did

Fungal infections

The notable increase in fungal infections among COVID-19 positive pneumonia patients is a significant finding. This aligns with emerging concerns about secondary infections in COVID-19 (Timsit *et al.*, 2018).[8] The association between COVID-19 and heightened susceptibility to fungal infections merits further exploration.

Comorbidities

The incidence of diabetes, hypertension, and COPD did not show significant differences between the two groups. This aligns with the multifaceted nature of COVID-19 outcomes, where comorbidities alone may not fully explain variations in severity (Guan *et al.*, 2020).[7]

Treatment modalities

Our study demonstrates a substantial disparity in the use of remdesivir and tocilizumab, highlighting potential differences in treatment approaches based on COVID-19 status. The higher use of remdesivir aligns with its established role in COVID-19 management.[9] The significant difference in tocilizumab use may reflect evolving clinical strategies for managing COVID-19 complications.

Invasive mechanical ventilation and duration of stay

The increased need for invasive mechanical ventilation and prolonged hospitalization in COVID-19 positive pneumonia patients is consistent with the heightened severity often associated with COVID-19.[10] These findings emphasize the critical impact of COVID-19 positivity on patient management and healthcare resource utilization.

Radiological and biomarker differences

Distinct radiological characteristics, as indicated by higher CT scores in COVID-19 positive pneumonia, align with existing knowledge of COVID-19's impact on lung imaging.[6] Elevated D-Dimer levels in COVID-19 positive pneumonia underscore the hypercoagulable state associated with severe COVID-19 cases.[8]

CRP levels

While not statistically significant, the trend towards elevated CRP levels in COVID-19 positive pneumonia is in line with studies highlighting the role of inflammatory markers in COVID-19 severity.[10]

Conclusion

The study presents a comprehensive analysis of asymptomatic fungal infections in ICU patients, comparing suspected and confirmed COVID-19 cases. The results reveal a noteworthy prevalence of fungal infections, emphasizing their significance in critically ill populations. The study shedsha light on potential variations between suspected and confirmed COVID-19 cases, contributing valuable insights for clinical practices and future research in COVID-19 management. Further investigation is warranted to enhance our understanding of these infections and refine patient care strategies in the challenging landscape of critical care.

Limitation of study

The limitations of this study include a modest sample size and a single-center design, which may impact the generalizability of the findings. Being a cross-sectional study, it provides a snapshot, limiting the ability to establish causal relationships and explore temporal changes. Retrospective data collection introduces the potential for selection bias and inconsistencies. Additionally, the evolving nature of COVID-19 knowledge and the lack of extended follow-up data constrain the study's comprehensive understanding. These limitations underscore the need for cautious interpretation and warrant consideration in future research.

Future directions

This study lays the groundwork for several avenues of future research aimed at enhancing our understanding of asymptomatic fungal infections in the context of COVID-19 pneumonia and critically ill patients. Firstly, longitudinal studies with larger sample sizes and multi-center designs could provide a more comprehensive assessment of fungal infection prevalence, risk factors, and outcomes over time. Additionally, exploring the impact of antifungal prophylaxis or early detection strategies on patient outcomes could inform guidelines for managing fungal infections in COVID-19 patients. Furthermore, investigating the role of host immune responses and genetic predispositions in fungal susceptibility may offer insights into personalized treatment approaches. Integrating advanced diagnostic techniques, such as metagenomic sequencing or biomarker profiling, may also enhance the early detection and characterization of fungal infections. Collaborative efforts between infectious disease specialists, pulmonologists, and microbiologists are essential to advance knowledge in this field and optimizing patient care strategies, particularly in the ever-evolving landscape of viral respiratory illnesses like COVID-19.

Author contributor

Residents doctors involved in CU care of patients.

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Conflicts of interest

There are no conflicts of interest.

References

1. Verweij PE, Rijnders BJA, Brüggemann RJM, Azoulay E, Bassetti M, Blot S, *et al.* Review of influenza-associated

pulmonary aspergillosis in ICU patients and proposal for a case definition: An expert opinion. Intensive Care Med 2020;46:1524-35.

2. Bartoletti M, Pascale R, Cricca M, Rinaldi M, Maccaro A, Bussini L, *et al.*; PREDICO Study Group. Epidemiology of invasive pulmonary aspergillosis among COVID-19 intubated patients: A prospective study. Clin Infect Dis 2020;73:e3606-14.

3. Koehler P, Cornely OA, Böttiger BW, Dusse F, Eichenauer DA, Fuchs F, *et al.* COVID-19 associated pulmonary aspergillosis. Mycoses 2020;63:528-34.

4. van Arkel ALE, Rijpstra TA, Belderbos HNA, van Wijngaarden P, Verweij PE, Bentvelsen RG. COVID-19-associated pulmonary aspergillosis. Am J Respir Crit Care Med 2020;202:132-5.

5. Garcia-Vidal C, Sanjuan G, Moreno-García E, Puerta-Alcalde P, Garcia-Pouton N, Chumbita M, *et al.* Incidence of co-infections and superinfections in hospitalized patients with COVID-19: A retrospective cohort study. Clin Microbiol Infect 2021;27:83-8.

6. Wu Z, McGoogan JM. Characteristics of and important lessons from the coronavirus disease 2019 (COVID-19) outbreak in China: Summary of a report of 72,314 cases from the Chinese center for disease control and prevention. JAMA 2020;323:1239-42.

7. Guan W-J, Liang W-H, Zhao Y, Liang H-R, Chen Z-S, Li Y-M, *et al.* Comorbidity and its impact on 1590 patients with COVID-19 in China: A nationwide analysis. Eur Respir J 2020;55:2000547.

8. Timsit J-F, Bassetti M, Cremer O, Daikos GL, de Waele JJ, Kaukonen KM, *et al.* New and emerging agents for the treatment of infections in the ICU. Critical Care. 2018;22:119.

9. Beigel JH, Tomashek KM, LE Dodd, Mehta AK, Zingman BS, Kalil AC, *et al.* Remdesivir for the treatment of Covid-19—Final report. N Engl J Med 2020;383:1813-26.

10. Richardson S, Hirsch JS, Narasimhan M, Crawford JM, McGinn T, Davidson KW, *et al.*; the Northwell COVID-19 Research Consortium. Presenting characteristics, comorbidities, and outcomes among 5700 patients hospitalized with COVID-19 in the New York City area. JAMA 2020;323:2052-9.