

Mucormycosis in patients infected with the delta strain of COVID-19 virus, effective of some factors in the prevalence and patient prognosis in Shahid Beheshti Hospital, Kashan, central Iran

Zahra Soleimani (MD) ¹
Kamal Esalatmanesh (MD) ²
Hossein Hooshyar (PhD) ³
Habibollah Rahimi (PhD) ⁴
Mojtaba Reisee (MD) ⁵
Roozbeh Esalatmanesh (MD) ^{6*}

1. Department of Infectious Diseases, Kashan University of Medical Sciences, Kashan, Iran
2. Department of Internal Medicine, Kashan University of Medical Sciences, Kashan, Iran
3. Department of Medical Parasitology, Kashan University of Medical Sciences, Kashan, Iran
4. Department of Biostatistics and Epidemiology, School of Public Health, Kashan University of Medical Sciences, Kashan, Iran
5. Medical student, Kashan University of Medical Sciences, Kashan, Iran
6. Department of Radiology, Kashan University of Medical Sciences, Kashan, Iran

* Correspondence:

Roozbeh Esalatmanesh,
Department of Radiology, Kashan University of Medical Sciences, Kashan, Iran

E-mail: esalatr@gmail.com

Tel: +983155540021

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Abstract

Background: Opportunistic infections such as mucormycosis, are spread in COVID-19 patients due to the use of corticosteroids therapy. This study aimed to determine the prevalence of mucormycosis in patients infected with the delta strain of the COVID-19 virus and evaluated some effective factors in the prevalence and patient prognosis.

Methods: This study was performed on 44 COVID patients with co-infection to mucormycosis who were admitted to Shahid Beheshti Hospital, Kashan in 2022. The COVID diagnosis was based on clinical signs and a positive PCR test, the diagnosis of mucormycosis was based on a positive laboratory report of fungi culture medium. The patient outcomes were recorded and CALL scoring system was used to determine the severity of the disease. The results were analyzed by descriptive statistics using SPSS 16 software.

Results: Of 44 COVID patients infected with mucormycosis, 26 (59.1%) had diabetes. All 44 patients had a history of taking steroids with an average daily dose of 48.77 ± 65.15 mg. Twenty-six (59.1%) patients were admitted to the ICU, 18 (40.9%) required intubation. Sixteen (36.4%) patients died. A significant decrease observed in lymphopenia after treatment of patients ($p < 0.0001$). CALL scores using showed that 33 (75%) patients had severe risk factors.

Conclusion: Uncontrolled diabetes and hypertension are the two most common risk factors of mucormycosis in COVID patients. High use of glucocorticoid drugs and immune suppression are predisposing causes for opportunistic organisms. Accurate laboratory diagnosis of mucormycosis plays an important role in treatment of patients and in controlling the disease.

Keywords: COVID-19, Mucormycosis, Immunosuppression, Delta strain, Corticosteroid therapy.

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Coronavirus disease which is also called the novel coronavirus (COVID-19) disease, was first reported in Wuhan, China, in 2019 (1-4). Human infection can be mild or asymptomatic, but many infections have mainly critical condition, leading to hospitalization and death (5-7). Common symptoms of this infection include fever, chills, fatigue, severe headache, cough, and shortness of breath or difficulty breathing in severe cases (8-9). In hospitalized patients, the most reported common symptoms included fever (up to 90%), dry cough (60 to 86%), shortness of breath (53 to 80%), fatigue (38%), nausea, vomiting, or diarrhea (15 to 39%) and myalgia (15 to 44%) (10). Pneumonia occurred in the second or third week of disease initiation. The pneumonia leads to interlobular involvement, alveolar secretions, and decreased oxygen saturation (11).



Lymphopenia is often accompanied by an increase in pro-inflammatory cytokines and inflammatory proteins such as C-reactive protein (CRP). Although mechanical ventilation is used for patients with pulmonary insufficiency, but base of treatment are oxygen therapy and symptomatic therapy. Acute respiratory distress, pneumonia, kidney failure, and death are seen in severe forms of coronavirus disease (11). Several laboratory finding, including high levels of D-dimer and severe lymphopenia, are related to critical illness, and mortality (3). Laboratory diagnosis is based on finding the Partial genome in samples taken from the pharynx and nose (RT-PCR), increasing PT, decreasing albumin and eosinophil, and increasing interleukin 6 (IL-6) levels (12). About 75% of symptomatic COVID-19 patients, can be detected by CT scans (13).

The delta variant of the COVID-19 virus (B.1.617.2) was first identified in India in May 2021 and was recognized as a concern strain of COVID-19 by the World Health Organization. Subsequently, this strain spread globally. A study in England showed that the transmission power of the delta strain was about 66% higher than the alpha type (14). Co-infection to a wide range of opportunistic bacterial and fungal agents is another consequence of this strain (15). *Aspergillus* spp. and *Candida albicans* have been reported as two main fungal agents causing co-infection in COVID-19 patients (16). Several cases of mucormycosis in COVID-19 patients were reported in certain regions of India, in 2020 spring, and then spread worldwide. Raut et al. reported that the spread of mucormycosis is one of the other challenges of the COVID-19 pandemic in India and recommended that the government public education and preventive measures should be taken to prevent deaths in COVID-19 patients (17). A review study by Singh et al. showed that the use of corticosteroids in the treatment of COVID-19 patients has increased the incidence of mucormycosis. Maintenance of the optimal level of blood sugar and the rational use of glucocorticoids in these patients could be effective in the prevention of co-infection (16). Mehta et al.'s study showed that high use of steroids, monoclonal antibodies, and broad-spectrum antibiotics may lead to the establish of mucormycosis, and physicians should be aware of the possibility of secondary invasive fungal infections in COVID-19 patients (18).

Considering the increasing importance of the COVID-19 pandemic and the spread of the delta strain, Increased use of glucocorticoids in the treatment of COVID-19, concurrent diabetes, and a higher probability of mucormycosis in these conditions, the present study was conducted was to determine the demographic, clinical, and

para-clinical findings, and outcomes of mucormycosis patients in hospitalized individuals infected with the delta strain of COVID-19 in Shahid Beheshti hospital, Kashan University of Medical Sciences.

Methods

This retrospective descriptive case study was performed on 44 COVID patients with co-infection to mucormycosis who were admitted to Shahid Beheshti Hospital in Kashan, central Iran, in 2022. The diagnosis of COVID-19 delta variant was based on clinical symptoms including fever, myalgia, sore throat, headache, cough, and shortness of breath, and a positive real-time polymerase chain reaction test (RT-PCR). Diagnosis of delta strain was based on the approval of the Comprehensive Health Laboratory of the Health Vice-Chancellor of Kashan University of Medical Sciences.

The diagnosis of mucormycosis infection has been based on clinical symptoms, including bloody secretions with tissue necrosis, especially in the nasal and sinus areas, and a positive laboratory report of fungi culture medium. The demographic, clinical, and laboratory findings, systemic diseases such as diabetes, consuming immunosuppressive drugs and glucocorticoids, and patient outcomes including hospitalization in Intensive Care Unit (ICU), ventilator use, and mortality were recorded. To determine the severity of the disease, the CALL scoring system was used, according to Ji et al. (19). Based on the CALL system, a score of 4 to 6 was considered as a good prognosis, a score of 7 to 9 as intermediate prognosis, and a score of 10 to 13 as a poor prognosis (19). Throughout the study, ethical considerations were observed and all patient information was kept confidential.

Ethics statement: This study was approved by the Ethics Committee of the Kashan University of Medical Sciences encoding (IR.KAUMS.MEDNT.REC.1400.121).

Statistical analysis: The collected data were analyzed using SPSS Version 16 software. Data were analyzed using chi-square test, Fisher's exact test and McNemar test. A $p < 0.05$ was considered significant.

Results

Totally, forty-four patients with laboratory-confirmed COVID-19 and mucormycosis were included in this study. Out of 44 studied cases, 23 individuals were males (52.3%), and 21 were female (47.7%). The average age of patients infected with mucormycosis was 62.80 ± 12.51 years old (age range: 29–87). Diabetes (59.1% including 50%

Diabetes type 2 and 9% Diabetes type 1), high blood pressure (52.3%), and heart disease (34.1%) were the most important background history observed in the majority of cases of mucormycosis in patients with COVID-19, followed by Hyperlipidemia (27.3%). Table 1 show the frequency of underlying disease of patients infected with COVID-19 and mucormycosis. The majority of patients with mucormycosis had cough (84.1%), weakness/lethargy (77.3%) fever (61.4%), body pain (54.5%), dyspnea (54.5%), headache (43.2%) and chill (43.2%). Other symptoms included sore throat, nausea and vomiting, abdominal pain, sweating, vertigo, anorexia and chest pain (table 2). The main oxygen saturation among patients with COVID and mucormycosis, was $87.48 \pm 7.79\%$ mm Hg. And the final oxygen saturation at the end of the treatment period was $90.80 \pm 8.36\%$ mm Hg. Comparison of C-reactive protein (CRP) and lactate dehydrogenase (LDH) levels during COVID-19 course showed in table 3. According to these findings, a significant difference was seen in lymphopenia before and after treatment ($p < 0.0001$). The CRP and LDH remained without significant change (table 3). An abnormality (moderate-to-severe COVID-19

pneumonia) were seen in chest CT scan among 43 of the participants (97.7%) and only one (2.27%) patient showed normal condition. The PCR test for COVID-19 has been positive in all patients. All of the patients were administered heparin and broad-spectrum antibiotics during the course of their hospitalization. Two (13%) patients had received favipiravir, and 37 (84.1%) patient's remdesivir as anti-viral treatment. All patients have received intravenous and/or oral corticosteroids ($n = 44$, 100%). The average dose of steroid therapy was 65.15 ± 48.77 mg. Insulin and/or oral antidiabetic drug were used only for diabetic subjects. Based on the scores of patients in the CALL system, scores of 11 to 13 indicate high-risk patients with a probability of more than 50% for disease progression. In the present study, 11, 12, and 13 CALL scores obtained by patients were seen in 20.5%, 4.5% and 47.7% of individuals, respectively. The lowest score with the frequency was 7 that seen for only one (2.3%) of patients (table 4). Final outcome of the patients, showed that the prognosis of 33 (75%) patients were high-risk, and 11 (25%) moderate-risk. Twenty-six (59.1%) patients were hospitalized in ICU, 18 (40.9%) patients required intubation and 16 (36.4%) patients died (table 5).

Table 1. Frequency of underlying disease of COVID-19 patients with mucormycosis hospitalized in Shahid Beheshti hospital, Kashan

History	Positive No (%)	Negative No (%)	Total No (%)
Heart disease	15 (34.1)	29 (65.9)	44 (100)
Hypertension	23 (52.3)	21 (47.7)	44 (100)
Hyperlipidemia	12 (27.3)	32 (72.7)	44 (100)
Hypothyroidism	4 (9.1)	40 (90.9)	44 (100)
Chronic kidney disease	2 (4.5)	42 (95.5)	44 (100)
Malignancy	2 (4.5)	42 (95.5)	44 (100)
Organ transplant history	1 (2.3)	43 (97.7)	44 (100)
Anemia	1 (2.3)	43 (97.7)	44 (100)
Behcet's disease	1 (2.3)	43 (97.7)	44 (100)
Bipolar	1 (2.3)	43 (97.7)	44 (100)
Fatty liver	1 (2.3)	43 (97.7)	44 (100)
Rheumatoid Arthritis	2 (4.5)	42 (95.5)	44 (100)
Single kidney	2 (4.9)	42 (95.5)	44 (100)
Diabetes: Type 1	4 (9.1)	40 (90.9)	44 (100)
Diabetes: Type 2	22 (50)	22 (50)	44 (100)

Table 2. The symptoms and clinical findings of the patients with mucormycosis

symptoms and clinical findings	Positive No(%)	Negative No (%)	Total No (%)
Fever	27 (61.4)	17 (38.6)	44 (100)
Body pain	24 (54.5)	20 (45.5)	44 (100)
Weakness and lethargy	34 (77.3)	10 (22.7)	44 (100)
Cough	37 (84.1)	7 (15.9)	44 (100)
Dyspnea	24 (54.5)	20 (45.5)	44 (100)
Sore throat	6 (13.6)	38 (86.4)	44 (100)
Diarrhea	5 (11.4)	39 (88.6)	44 (100)
Headache	19 (43.2)	25 (56.8)	44 (100)
Nausea and vomiting	16 (36.4)	28 (63.6)	44 (100)
Chill	19 (43.2)	25 (56.8)	44 (100)
Sweating	1 (2.3)	43 (97.7)	44 (100)
Abdominal pain	1 (2.3)	43 (97.7)	44 (100)
Vertigo	5 (11.4)	39 (88.6)	44 (100)
Anorexia	11 (25)	33 (75)	44 (100)
Chest pain	7 (15.9)	37 (84.1)	44 (100)

Table 3. Comparison of laboratory findings in COVID-19 patients before and after treatment

Laboratory finding			After treatment		P- Value
			Positive NO (%)	Negative NO (%)	
Lymphopenia	Before treatment	Positive	25 (45.4)	19 (43.2)	0.0001
		Negative	0	5 (11.4)	
CRP	Before treatment	Positive	13 (29.5)	16 (36.4)	0.093
		Negative	7 (15.9)	8 (18.2)	
High LDH	Before treatment	Positive	8 (50)	6 (37.5)	0.289
		Negative	2 (12.5)	0	

Table 4. The scores of 44 COVID-19 patients with mucormycosis in the CALL system

Scoring	No	(%)
7	1	2.3
8	3	6.8
9	6	13.6
10	2	4.5
11	9	20.5
12	2	4.5
13	21	47.7
Total	44	100

Table 5. Final outcome of the 44 COVID-19 patients with mucormycosis in Shahid Beheshti Hospital, Kashan

Outcome	Positive No (%)	Negative No (%)	Total No (%)
Death	16 (36.4)	28 (63.6)	44 (100)
Hospitalization in ICU	26 (59.1)	18 (40.9)	44 (100)
Intubation	18 (40.9)	26 (59.1)	44 (100)

Discussion

COVID-19 is associated with a wide range of opportunistic bacterial and fungal infections (17). It seems that the main cause of facilitating the occurrence of fungal diseases in patients with COVID-19, is the proper environment for growth of fungi organisms in these patients due to hypoxia, high glucose (especially in diabetic patients), metabolic acidosis, and diabetic ketoacidosis. A decrease of the phagocytic activity of white blood cells, immunosuppression (due to SARS-CoV-2, or due to the use of steroids to reduce inflammatory reactions in COVID-19 patients) and prolonged hospitalization duration with or without mechanical ventilator in patients are the other common risk factors (20, 21). According our findings, uncontrolled diabetes is the most underlying disease in mucormycosis. Correct and rapid diagnosis and correct treatment, play a very important role in the prognosis of the disease and reduce the mortality of patients.

Control or treatment of the underlying disease in patients with COVID-19 and diabetes is very important. Bari et al.'s study (2021) showed that mucormycosis was detected in 85.5% of cases of COVID-19 delta strain and 64.11% of diabetic patients in South Asian regions (22). According to the present study, diabetes, high blood pressure, and heart disease were the most common underlying diseases among mucormycose patients with COVID-19, respectively. Mahalaxmi et al.'s study showed that the most of mucormycose patients had diabetes during the outbreak of the delta strain of COVID-19 in 2021 (23). Jeong et al. reported that diabetes and immunosuppression are the main risk factor of mucormycosis (24). A study conducted by Ge Song et al. Showed that patients with immune system deficiency, are more likely to be infected with invasive fungi (16). A survey on 9 patients with mucormycosis, showed that all of these patients had diabetes or immune system deficiency (25).

In the present study, all patients received heparin and antibiotics, and most of them received hydrocortisone and remdesivir. All 44 patients used steroids and their average daily dose was 65.15±48.77 mg. In Mahalaxmi et al.'s study, the most of the patients had diabetes, and steroids were prescribed for them (23). Study of Singh et al., showed

that the use of corticosteroids in the disease of COVID-19 increase the possibility of mucormycosis. It was also emphasized that in these patients, all efforts should be made to maintain the optimal level of glucose and rational use of corticosteroids (15). A review study in 2021, showed that immunosuppressive treatment in COVID-19 patients, leads to them suffering from opportunistic infections (26). Salil Mehta et al. showed that the widespread use of steroids, monoclonal antibodies, and broad-spectrum antibiotics may lead to the development or exacerbation of fungal disease (18). In the present study, the most common symptoms among the patients were cough, weakness and lethargy, fever, and shortness of breath, respectively. Complaints such as loss of smell and taste were not observed among the patients. Al-Tawfiq et al.'s study showed that the most common symptoms in patients with COVID-19 and mucormycosis are pulmonary manifestations including shortness of breath and cough, and subsequently, skin and mucosal manifestations (22).

Colds and sinusitis, and gradually developed blindness, proptosis, paralysis of the fifth and seventh cerebral nerves, and decreased level of consciousness were the symptoms of 9 patients with mucor that reported by Mohammad et al.'s study (25). It is reported that the first complaints of patients with COVID-19 and mucormycosis were related to intranasal symptoms, include hyperemia and nasal obstruction which are usually non-specific. Diagnosis at this stage requires strong clinical suspicion. In high-risk individuals, resistant rhinitis may be the first sign of involvement with mucormycosis (27). The most common para-clinical findings among the patients in present study were abnormal lung CT scan, lymphopenia, high LDH, abnormal radiography and high CRP in the beginning of hospitalization respectively. In a similar study, abnormal CT scans, and high CRP were the most common para-clinical findings (22). In a case report by Moeini and Mehraban, leukocytosis, high ESR, high CRP titer, and infiltration in the middle lobe of the right lung was observed in a patient infected with pulmonary mucormycosis and COVID-19 (28). Final outcome of the patients, in the present study showed that the prognosis of 75% patients were high-risk, and 25% moderate-risk. Twenty-six

(59.1%) patients were hospitalized in ICU, and 16 (36.4%) patients died. A review study conducted by Tawfiq in 2021 showed that the death rate due to this disease was 28%. However, mortality disease, in the presence of diabetes, was 49% (21). Jeong et al.'s study estimated the mortality rate of COVID-19 patients due to mucormycosis is 45% (24). It seemed that the difference between the mortality rates of COVID-19 patients with mucormycosis in different studies, related to health status of patients. In the present study, the highest score obtained by the patients was 13 (47.7%) and 11 (20.5%), respectively, which indicates the impact of comorbidity (combined diseases, age over 60 years) and other risk factors. To the best of our knowledge, the present study is the first report that evaluates the prognosis of COVID-19 patients with mucormycosis based on the CALL system. However, further studies are needed and recommended.

According to the findings of this study, uncontrolled diabetes and high blood pressure are the most common risk factors for mucormycosis. Use high doses of steroids among COVID-19 patients and the findings of previous studies that considered immune system suppression as a predisposing factor for opportunistic infections, Mucormycosis can be related to steroid treatment in these patients. Considering the effect of comorbidity in contracting mucormycosis in patients with COVID-19 and also high mortality of these patients, correct and rapid diagnosis and correct treatment play a very important role in the prognosis of the disease and reduce the mortality of patients significantly.

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Authors' contribution: ZM and MR designed the conception of the study; HH and KE focus of the statically analysis; ZM and RE technical support and conceptual advice. All authors contributed to the draft of the manuscript, revised it critically, and approved the final version.

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