MÆDICA - a Journal of Clinical Medicine

CASE REPORT

MAEDICA – a Journal of Clinical Medicine
2026; 21(2): 532-538
https://doi.org/10.26574/maedica.2026.21.2.532

Subglottic Mucormycosis and Invasive Candidiasis in Uncontrolled Diabetes Mellitus – a Case Report with Review of the Literature

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ABSTRACT

Mucormycosis is an opportunistic infection caused by fungi of the order Mucorales and Candida *is a yeast which is the most common cause of fungal infections in humans. The most commonly known risk factor for these fungal infections is uncontrolled diabetes mellitus (DM), followed by other causes of immunosuppression like neutropenia and corticosteroid therapy. In atypical clinical presentations, all differential diagnosis should be considered, and followed by histopathological and microbiological examination for diagnosis of fungal infections like mucormycosis and candidiasis in uncommon locations. Early diagnosis and combined medical and surgical treatment, along with resolution of the associated risk factors can lead to effective results and good prognosis.*

Keywords: candidemia, diabetic ketoacidosis, immunosuppression, laryngeal mucormycosis, subglottic region, vocal cord palsy.

ABBREVIATIONS

DM: diabetes mellitus
ROCM: rhino–orbito–cerebral mass
COVID-19: coronavirus disease 2019
DKA: diabetic ketoacidosis
CT: computed tomography
CBNAAT: Cartridge-Based Nucleic Acid Amplification test
PWD: people with DM
IFN-γ: interferon-γ
HIV: human immunodeficiency virus
MALDI-TOF MS: matrix-assisted laser desorption ionization time-of-flight mass spectrometry
IDSA: Infectious Diseases Society of America

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Article received on the 14th of October 2025 and accepted for publication on the 19th of November 2025

INTRODUCTION

Mucormycosis is an uncommon opportunistic infection caused by fungi of the order Mucorales (1). It most commonly presents as a rhino–orbito–cerebral mass (ROCM) especially in people with diabetes mellitus (DM). Rarely, in post-transplant recipients and cases of haematological malignancies, pulmonary mucormycosis can occur (2). The paranasal sinuses are commonly involved; howfever, the laryngeal tract involvement is very rarely seen (3). The most common risk factor is uncontrolled DM, followed by other immunosuppressive conditions like neutropenia, corticosteroid therapy and coronavirus disease 2019 (COVID-19) (3, 4). Uncontrolled DM and hyperglycemia lead to immune dysfunction, due to which the patient becomes prone to many bacterial and fungal infections. Invasive fungal infections whether community acquired or hospital acquired are more common in people with uncontrolled DM. The risk of candidiasis also increases in uncontrolled DM, which may present as superficial cutaneous or mucocutaneous infections or invasive candidiasis (5). Hereby we describe a case of subglottic mucormycosis and invasive candidiasis in a patient with diabetic ketoacidosis (DKA) who presented with dyspneoa and septicemia. The patient underwent emergency tracheostomy, followed by bronchoscopy. Initially, there was a suspicion of malignancy; however, the final diagnosis revealed mucormycosis with candidiasis. ❑

CASE REPORT

A 56-year-old female patient presented with generalised weakness as well as difficulty breathing and swallowing for 15 days. On examination, there was pallor and tachycardia. Bronchial breath sound was present on the left side. The patient had anorexia for two days and constipation for five days. She was a known case of DM on treatment for the last 10 years and was recently admitted in another hospital for one week due to DKA and septicemia. Computed tomography (CT) thorax was performed, which showed fibroatelectatic opacities in the middle and lingual lobe with tractional bronchiectasis. Subtle fibronodular opacities were also seen in the right lower lobe.

Investigations and treatment

Various investigations were performed for effective diagnosis and management of the patient (Table 1). On biochemical evaluation, her random blood glucose reading was 541 mg/dL and

Sequential timeline		Performed test	Result (normal lab reference range)
1	On admission	Random blood glucose	541 mg/dL (70-140 mg/dL)
		HbA_{1c}	14.9 % (<5.7%)
		Potassium	2.8 mEq/L (3.5 to 5.0 mEq/L)
		Haemoglobin	11.8 g/dL
		Total leucocyte count	31550/uL (4000-11000/uL)
		Platelet count	6,74,000/uL(1,50,000- 4,00,000/uL)
2		C-reactive protein	140.1 mg/L (3 mg/L)
3		Erythrocyte sedimentation rate	75 mm/Hr (<30 mm/Hr)
4		Bronchoscopy	Right true vocal cord was fixed on adduction, phonation gap was present and there was subglottic congestion with whitish areas. There was right sided vocal cord palsy
5		Bronchial biopsy for microbiological examination	Presence of pseudomonas and candida infection on microbiological culture
6		Blood culture	Candidemia
7		Subglottic biopsy for histopathological examination	Mucormycosis with candidiasis
8		Biopsy from gastric antrum biopsy	Superficial chronic gastritis
9		Pleural fluid cytology	Inflammatory. No granulomas/ organisms/malignancy
10		CT scan of neck and thorax	An ill-defined soft tissue density lesion with non-enhancing necrotic areas within of size 78 x 48 x 63 mm was noted in the right middle lobe of lung with possibility of an infective lesion more than a neoplastic lesion.

TABLE 1. Sequential order of investigations performed for patient management

HbA_{1c} 14.9% along with hypokalemia (Potassium 2.8 mEq/L). Her haemoglobin was 11.8 g/dL, the total leucocyte count was 31550/uL with neutrophilic leucocytosis and platelet count of 6,74,000/uL. Inflammatory markers like C-reactive protein (CRP) (140.1 mg/L) and erythrocyte sedimentation rate (75 mm/hour) were elevated. Emergency tracheostomy was done. Bronchoscopy was performed wherein the right true vocal cord was fixed on adduction, phonation gap was present and there was subglottic congestion with whitish areas (Figure 1). There was right sided vocal cord palsy. Clinically there was a suspicion of malignancy. A bronchial biopsy was sent for microbiological examination and a subglottic biopsy was sent for histopathological examination.

Tracheobronchial tissue showed the presence of *Pseudomonas* and *Candida* infection on microbiological culture. Blood culture showed the presence of candidemia. Tracheal-bronchial tissue culture identified pseudomonas sensitive to piperacillin, ceftizidine, ciprofloxacin, levofloxacin. *Candida krusei* (current name *Pichia kudriavzevii*) was isolated on blood culture which was sensitive to caspofungin, micofungin, amphotericin B, voriconazole and resistant to fluconazole.

The subglottic biopsy revealed broad, non-septate fungal hyphae with 90-degree angle branching, characteristic of *Mucor*, along with neutrophils and presence of yeast and pseudohyphae forms of *Candida* infiltrating the respiratory epithelium and vessels in the stroma (Figure 2). This was confirmed by Periodic acid-Schiff and Grocott methenamine silver stains (Figures 3 and 4). There was no evidence of dysplasia or malignancy. Findings were suggestive of mucormycosis with candidiasis. The culture was not done for *Mucor*; therefore, the exact identification of the involved fungus at the genus and species level was not possible.

Biopsy from gastric antrum biopsy was also sent, which revealed superficial chronic gastritis. Pleural fluid was sent for cytology, which showed inflammatory cells with lymphocytic predominance, few neutrophils and macrophages. No organisms or granulomas were identified. Tuberculosis was not detected on bronchial wash

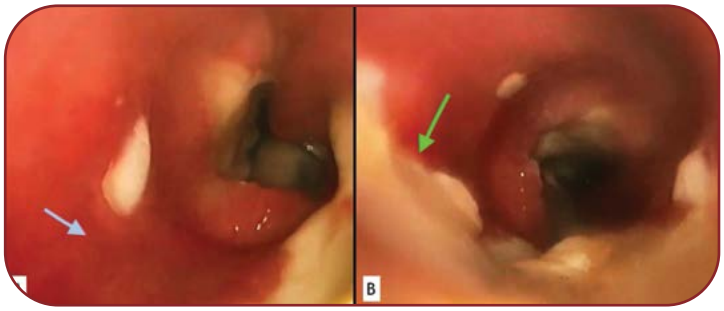

FIGURE 1. Bronchoscopy showing features clinically suspicious of malignancy: A) widespread subglottic congestion (blue arrow) with focal whitish areas; and B) many whitish patchy areas (green arrow)

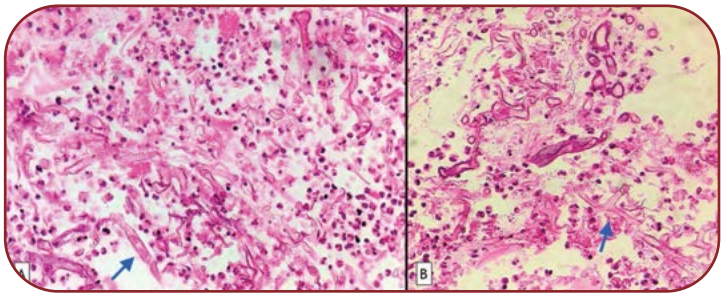

FIGURE 2. Subglottic biopsy showing A) broad non-septate hyphae of *Mucor* and B) having 90-degree angle branching (Haematoxylin and Eosin stain, 400x magnification)

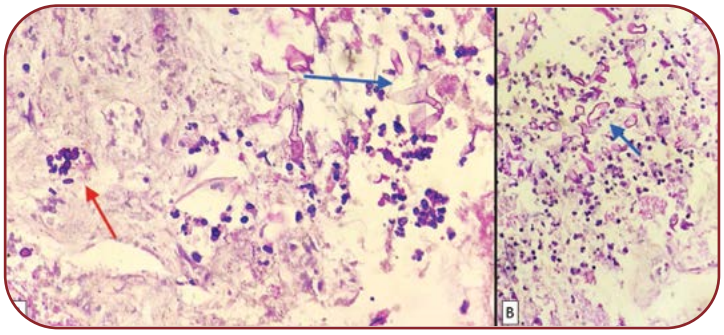

FIGURE 3. Periodic acid-Schiff stain highlighting A) aseptate, broad, hyphae of Mucor (blue arrow) along with candidal spores (red arrow) and B) broad, non-septate hyphae of *Mucor* (400x magnification)

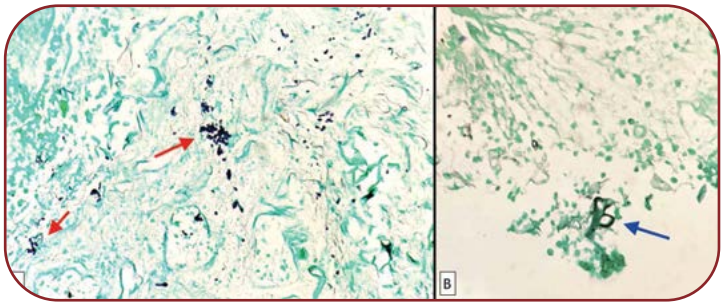

FIGURE 4. Grocott methenamine silver stain highlighting A) candidal spores (red arrow) and B) broad non-septate hyphae of *Mucor* (blue arrow) (400x magnification)

Cartridge-Based Nucleic Acid Amplification test (CBNAAT).

Thereafter, a CT scan of neck and thorax was done, which revealed the presence of diffuse thickening of vocal cord on either side causing narrowing of laryngeal inlet. Bilateral moderate pleural effusion was noted, with segmental collapse of both lower lobes of the lung. An ill-defined soft tissue density lesion with non-enhancing necrotic areas within of size 78 x 48 x 63 mm was noted in the right middle lobe of the lung. Radiologically possibility of an infective lesion was suggested more than a neoplastic lesion.

Outcome

The patient was diagnosed as a case of DKA with hypokalemia, right sided vocal cord palsy, *Pseudomonas* and candidiasis in the tracheobronchial tissue, candidemia and mucormycosis with candidiasis in subglottic region. The patient was treated with regular insulin, glargine, liposomal amphotericin B (200 mg/day), meropenem and pantoprazole continued for one month based on current guidelines (3). The surgical debridement could not be carried out due to inaccessibility of the affected area by a general surgeon, presence of multiple co-morbidities, unavailability of a specialized cardiothoracic vascular surgeon and the family's reluctance to undergo a surgical intervention. Even after extensive medical treatment for a month, the condition of the patient deteriorated and she was kept on assisted ventilation but finally succumbed to death after 45 days of admission. ❑

DISCUSSION AND REVIEW OF THE LITERATURE

Diabetes mellitus is one of the most prevalent non-communicable diseases in India. According to the International Diabetes Federation data of 2021, 537 million people between the age of 20-79 years are living with DM globally, while the prevalence of DM in India is 8.7%. The risk of mycoses in people with DM (PWD) increases to 1.38 folds compared to non-diabetics (5). Invasive fungal infection common in DM are candidiasis, cryptococcosis, aspergillosis, mucormycosis primarily with *Rhizopus* spp. and fusariosis (5).

In hyperglycemic conditions, nicotinamide adenine dinucleotide phosphate hydrogen is intensively used for polyol pathway, which impairs the recycling of glutathione and decreases its level. Increased glucose also raises the level of reactive oxygen species and pro-inflammatory cytokines in blood, which in turn limits the activity of macrophages. The ability of the immune cells to secrete various cytokines like Interleukins 1, 6, 10, 12, 17A and interferon-γ (IFN-γ) is decreased, preventing containment of infection. Phagocytosis is defective due to impaired activity of complement receptors on monocytes. Hyperglycemia in DM impairs the degranulation of neutrophils, which in turn leads to an increased likelihood of infection and reduced clearance of pathogen, leading to invasive disease (5).

Invasive fungal infections in diabetes mellitus

1. Mucormycosis

Mucormycosis is an opportunistic angio-invasive fungal infection arising due to saprophytic fungi of the order *Mucorales*. Due to absence of population-based studies, the incidence of mucormycosis in India is not known but the estimated prevalence is almost 70 times higher than that reported throughout the world (6). There was an increase of mucormycosis cases during the COVID-19 pandemics (7). Mortality due to mucormycosis is 70-80% in diabetic people compared to overall mortality of 40-50% (8, 9). In a resource limited setting like India, many PWD do not seek treatment until complications begin and hence are diagnosed late or they present in DKA. In one study, mucormycosis was diagnosed in 74% of people with uncontrolled hyperglycemia, out of whom 43% were presenting for the first time with hyperglycaemia identified as newly diagnosed DM (5).

Based on the site of involvement, mucormycosis can present as a ROCM, pulmonary, gastrointestinal, cutaneous lesion, disseminated mucormycosis or can have uncommon presentations like renal involvement, endocarditis, osteomyelitis and pericarditis. Immunosuppression is the most important risk factor including cases having DM, haematological malignancy, extensive skin injury, human immunodeficiency virus (HIV) in-

fection, iron overload, history of voriconazole therapy, chemotherapy, peritoneal dialysis, post-haematopoietic stem cells and solid-organ transplant recipients on immunosuppressive therapy. However, mucormycosis has been reported in immunocompetent hosts too (6).

People with uncontrolled DM are more likely to present with ROCM and 67% of ROCM cases occurs in PWD. It may present with eye and facial swelling, proptosis, headache, visual disturbances and sudden vision loss (10). Pulmonary mucormycosis is not common in DM but when it occurs it presents with significant complications like endotracheal involvement, thickening of the posterior tracheal band, vocal cord palsy, atelectasis, cavitary lesion, effusion and mediastinal or hilar lymphadenopathy (11).

We report a rare case of subglottic mucormycosis in a patient with DKA which was associated with vocal cord palsy. There are only a few case reports of subglottic mucormycosis and its prevalence is unknown (3, 12-15). Amirzagar *et al* documented a case involving a 51-year-old female patient diagnosed with COVID-19, who exhibited respiratory distress and was found to have a subglottic lesion, later identified as subglottic mucormycosis through histopathological analysis. The treatment regimen included surgical debridement, dexamethasone, liposomal amphotericin B and oral Posaconazole. After a period of four weeks, the patient's overall health significantly improved and there were no signs of recurrence during subsequent outpatient follow-ups (3). Mohindra *et al* reported on a 40-year-old female with diabetes who developed tracheal mucormycosis following H1N1 influenza. Her treatment involved amphotericin B and insulin injections, followed by surgical debridement. The patient showed recovery at the four-month follow-up (14). Wolf *et al* described a 61-year-old male with poorly controlled diabetes who presented with intraluminal mucormycosis affecting the first tracheal ring. The infected tissue was surgically resected and the patient received extended treatment with liposomal amphotericin B. He made a full recovery and was discharged in good health two months later (15). Surgical debridement was not possible due to inaccessible location. In this particular case, the patient was administered regular insulin, glargine, liposomal amphotericin B at a daily dose of 200 mg, meropenem and pantoprazole for one month. Due to the inaccessibility of the affected area by the general surgeon, presence of many comorbidities, non-availability of specialised cardiothoracic vascular surgeon and reluctance of the patient's family for surgical treatment, surgical debridement could not be performed. As the patient's health declined, she required assisted ventilation, but unfortunately, she succumbed to her condition 45 days following her admission.

2. Candidiasis

Candida, a yeast that is the main contributor of fungal infection in humans, may manifest as superficial cutaneous or mucocutaneous infections or invasive candidiasis in the form of candidemia, peritonitis, endophthalmitis, endocarditis and disseminated candidiasis. 90% cases of fungemia are due to *Candida* species with mortality ranging from 40% to 80%. Most of the studies on invasive candidiasis show candidemia with or without localisation (16).

The risk factors for candidemia include broad spectrum antibiotic therapy, steroid use, uncontrolled diabetes, parenteral nutrition, intravascular catheters and gastrointestinal mucosal damage. In past few decades, there has been a shift in the *Candida* species causing the blood stream infections from *C. albicans* to non-albicans *Candida* (16). In this case, patient with uncontrolled DM developed mucocutaneous candidiasis along with invasive candidiasis in the form of candidemia and *Candida krusei* was the isolated.

Diagnosis and treatment of invasive fungal infections

1. Mucormycosis

Early diagnosis and defining the extent of invasion is important for successful management of mucormycosis. Endoscopic examination of sinuses, imaging, microbiological and histopathological examination of the proper sample forms the mainstay of diagnosis. Endoscopy of sinuses provide the early indication of invasion. Radiological imaging of paranasal sinuses and other

involved areas provide information regarding extent of invasion (17).

For laboratory diagnosis, nasal discharge or excised tissue is sent for potassium hydroxide mount and culture. Direct microscopy in particular, is a rapid, sensitive, reliable and inexpensive method for prompt diagnosis of mucormycosis. Culture is important for the exact identification of the fungus and the performance of susceptibility testing when necessary. Histopathological examination of tissue biopsy, as well as direct microscopy, will show characteristic broad non-septate 90-degree angle branching fungal hyphae (17). In this case, there was no evidence of a lesion in the sinus, no nasal discharge was observed, and the initial CT scan did not demonstrate any invasive mucormycosis, yet there was a clinical suspicion of malignancy. Histopathological examination of subglottic biopsy diagnosed mucormycosis.

Numerous disorders can manifest as a necrotic lesion in the subglottis both clinically and radiologically. These differential diagnoses include neoplasms such as chondrosarcoma, adenoid cystic carcinoma and lymphoproliferative disorders; infectious lesions such as bacterial, tuberculosis, aspergillosis and mucormycosis; inflammatory conditions such as Wegener's granulomatosis and sarcoidosis; and secondary tracheal wall ischemic injury brought on by prolonged intubation. In these situations, a biopsy is necessary for a precise diagnosis and treatment (15).

According to the current guidelines, a combined medical and surgical treatment is recommended for the best outcome (3). The main antifungal drug for mucormycosis is liposomal amphotericin B. Additionally, surgical debridement of the involved area is also required. Various modalities to improve immune functions like administrating granulocyte macrophage colony-stimulating factor to improve quantity and function of neutrophils, giving IFN-γ to enhance the function of monocytes, macrophages and neutrophils are also used (3, 14-15).

The mortality rate for subglottic mucormycosis is reported to be high. However, patient outcome and prognosis, including the mortality rate and survival, is dependent on several factors like the severity of underlying diseases, early incorporation of anti-fungal treatment and effective surgical debridement. Cases of tracheal mucormycosis having limited pulmonary involvement have better patient outcome and prognosis (3). If the immunocompromised state is not treated, even surgical debridement may not suffice and could require repetition, potentially progressing to disseminated disease. On the other hand, according to a study, effectively controlling underlying DM and antifungal therapy even without surgical debridement in localized diseases is an effective management strategy. Hence, the underlying immunosuppressed condition should also be vigorously managed and unchecked use of corticosteroid should be stopped as mucormycosis is uncommon in immunocompetent people (13). In our case, surgical debridement was not possible, so the patient was kept on liposomal amphotericin B and meropenem.

3. Candidemia

On tissue biopsy, *Candida* can be identified as spores and pseudohyphae forms of fungi. Blood culture is gold standard for the diagnosis of candidemia, which has been made easy by availability of automated identification systems and matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) that, despite a low sensitivity ranging from 21-71%, it has an advantage of species identification and antifungal susceptibility (18). Among the non-cultural methods, β-D-glucan is one the widely used test to detect the presence of this fungal cell wall component in a patient's blood serum but is also found in other fungi like *Aspergillus*. It has 92% sensitivity and 81% specificity. It has good negative predictive value to exclude the infection and stop the unnecessary treatment timely (18).

Infectious Diseases Society of America (IDSA) recommends echinocandins as the initial therapy for treating candidemia, followed by switching to azoles after 5-7 days, depending upon the susceptibility of the isolate, patient's stability and negative fungal blood culture. Duration of therapy is usually 14 days after the blood culture becomes negative (19)

The limitation of this case study was that facility for mucormycosis culture testing was not available in our centre. ❑

CONCLUSION

Hence, in a case with clinical and radiological presentation as a subglottic mass or lesion, all differential diagnosis should be clinically considered. Histopathological and microbiological examination is the cornerstone for identifying and diagnosing fungal infections like mucormycosis and candidiasis in uncommon locations. Early diagnosis and combined antifungal therapy and surgical debridement of necrotic tissues along with prompt and effective blood glucose control are the key to successful outcome. ❑

Authors' contributions: Garima Anandani – writing the original draft, review and editing; Anil Chaudhary, Parth Goswami – data curation, investigation; Mayuri Bhise, Komal Kumar Jangir, Kunal Deokar – data curation, project administration, supervision; Ashwini Agarwal – data curation, investigation, writing the original draft, review and editing.

Data availability: Data has already been shared in the present case report. Only patient's identity is not revealed, which is available on request.

Ethical statement: The authors confirm their adherence to the ethical policies of the journal. No identification details of the patient are shared.

Patient informed consent: Written informed consent for publishing this report in accordance with the journal's patient consent policy, before demise, was obtained from the patient.

Conflicts of interest: none declared.

Financial support: none declared.

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