International Journal of Research in Medical Sciences
Jaju MR et al. Int J Res Med Sci. 2020 Jun;8(6):2284-2296
www.msjonline.org
pISSN 2320-6071 | eISSN 2320-6012

DOI: http://dx.doi.org/10.18203/2320-6012.ijrms20202281

Case Series

Different types of mucormycosis: case series

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Received: 08 February 2020
Revised: 16 April 2020
Accepted: 22 April 2020

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ABSTRACT

Mucormycosis is the third invasive mycosis in order of importance after candidiasis and aspergillosis and is caused by fungi of the class Zygomycetes. The most important species causing Mucormycosis is Rhizopus arrhizus (oryzae). Identification of the agents responsible for mucormycosis is based on macroscopic and microscopic morphological criteria, carbohydrate assimilation and the maximum temperature compatible with its growth. The incidence of mucormycosis is approximately 1.7 cases per 1000 000 inhabitants per year. Clinical diagnosis of mucormycosis is difficult, and is often made at a late stage of the disease or post-mortem. We present here a series of five cases of different types of mucormycosis that were reported in our hospital till date. Of which three patients had good recovery and other two had a fatal outcome. Treatment of mucormycosis requires a rapid diagnosis, correction of predisposing factors, surgical resection or debridement as part of source control-and appropriate anti-fungal therapy. Liposomal amphotericin B is the drug of choice for this condition. The overall rate of mortality of mucormycosis is approximately 40%.

Keywords: Amphotericin-B, Mucorales, Mucormycosis, Rhizopus, Zygomycetes

INTRODUCTION

Mucormycosis, the third invasive mycosis in order of importance after candidiasis and aspergillosis, is a disease caused by fungi of the class Zygomycetes. The term 'mucormycosis' is used throughout this review of infections caused by Mucorales. The class Zygomycetes is divided into two orders, Mucorales and Entomophthorales. Members of the order Mucorales are the aetiological agents of the disease traditionally known as 'mucormycosis', a fulminant disease with high rates of morbidity and mortality that mainly affects immunocompromised patients. However, species of the order Entomophthorales are responsible for the chronic subcutaneous disease observed in immunocompetent patients in tropical and sub-tropical regions.[1]

Epidemiology

The incidence of mucormycosis is approximately 1.7 cases per 1000 000 inhabitants per year, which means 500 patients per year in the USA.[2] The main risk-factors for the development of mucormycosis are ketoacidosis (diabetic or other), iatrogenic immunosuppression, especially when associated with neutropenia and graft vs. host disease in haematological patients, use of corticosteroids or deferoxamine, disruption of mucocutaneous barriers by catheters and other devices,

and even exposure to bandages contaminated by these fungi.[3-9] This increase has generally taken place in patients and units where broad-spectrum antifungal prophylaxis, especially voriconazole, is used against Aspergillus.[10-12]

CASE PRESENTATION

Case 1

A 75-year-old male patient of uncontrolled diabetes mellitus with RBS 440mg/dL and nil for ketones in urine was admitted for blockage of nose on left side with swelling. He was diagnosed for left maxillary sinusitis and had undergone FESS. On the procedure he had blackish discharge for which the specimen was sent for microbiology and histopathology examination. The HPE showed chronic inflammation and single necrotic tissue bit with fungal hyphae with morphological features positive for mucormycosis with zygomycetes species. Later orbital cellulitis secondary to Mucormycosis was suspected. After FESS, the patient was given I.V. AMPHOTERICIN-B infusion for 60 days. Patients postoperative period was uneventful. Patient showed improvement and was discharged in stable condition with medical advice of taking tab. posaconazole.

In this case, the patient had left maxillary sinus mucormycosis (Figure 1-3) which responded well to FESS and Antifungals.

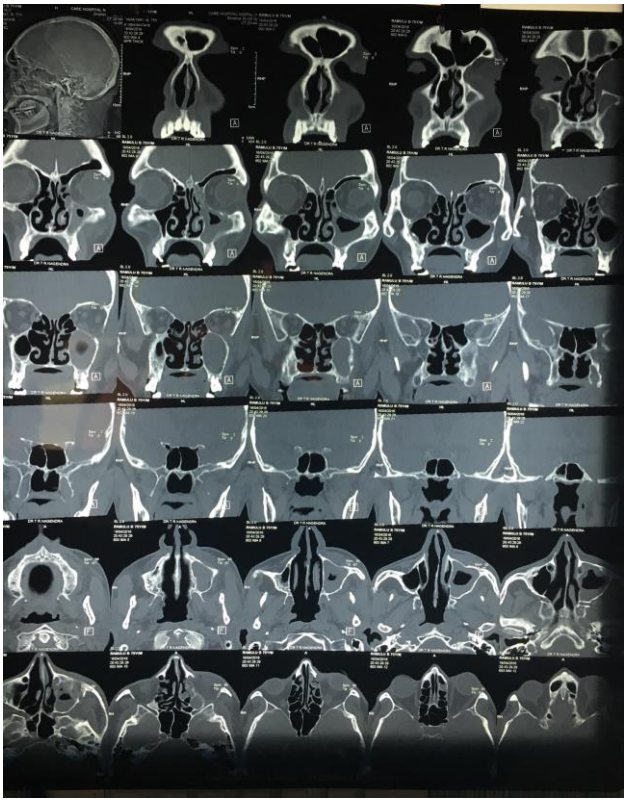

Figure 1: CT Scan maxillary sinusitis.

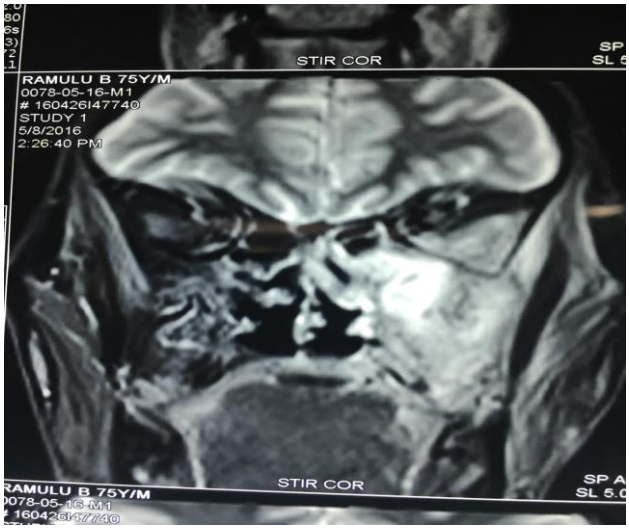

Figure 2: MRI scan diffuse sinusitis.

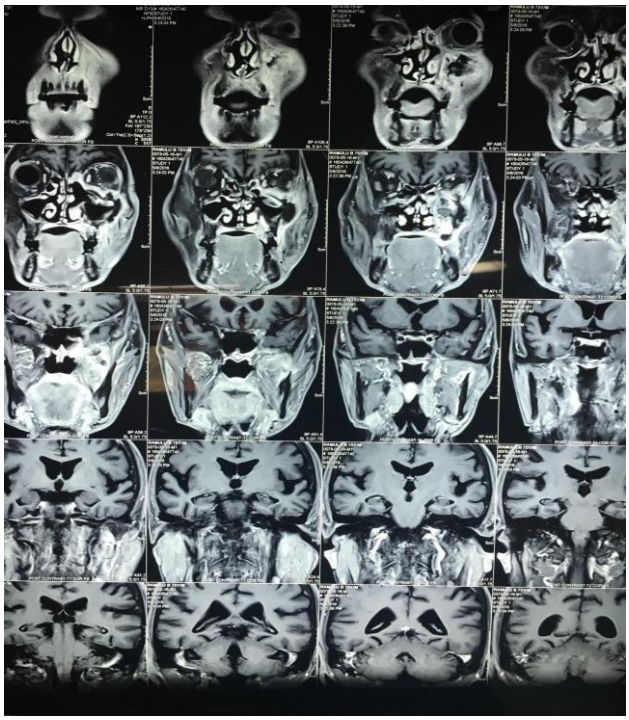

Figure 3: MRI scan different segments suggestive of diffuse sinusitis.

Case 2

A 41-year-old female patient known case of uncontrolled diabetes with RBS 400mg/dL and nil for ketones in urine, hypertensive presented with history of bleeding from right nostril, decreased vision in right eye, swelling and decreased sensation over right half of face for three days. On examination, she was conscious with stable vitals. On clinical examination, there was proptosis on right side, redness and complete ophthalmoplegia of right eye. Corneal reflex of right eye was absent. CT-PNS showed maxillary and ethmoid sinusitis. MRI Brain showed orbital cellulitis with optic nerve involvement. On ENT

opinion FESS was done. The samples were sent for HPE and fungal staining and culture. HPE showed fungal infection with organisms' morphology positive for mucormycosis with zygomycetes species. Blood for fungal culture-sensitivity and galactomannan assay came positive for aspergillus. Her orbital cellulitis persisted. Ophthalmologist opinion was taken who initially opted for conservative management as there was perception of light. Repeated MRI Brain with orbital cuts was suggestive of orbital cellulitis with optic nerve involvement and cavernous sinus involvement with cerebritis. Later her perception of light was also lost, her general conditions deteriorating, culminating into sepsis. After review with both ophthalmologist and neurosurgeon, exenteration of right eye and removal of intranuclear part of optic nerve was done under GA followed by local irrigation of eye socket with amphotericin along with systemic antifungals. She gradually improved over next few days, was extubated. She was discharged on taking Inj. amphotericin-B and Inj. caspofungin.

In this case, the patient had rhino-orbito-cerebral mucormycosis which responded to multiprong approach including FESS, eye exenteration (Figure 4) followed by irrigation of cavity with infusion of AMPHOTERICIN-B and systemic antifungals.

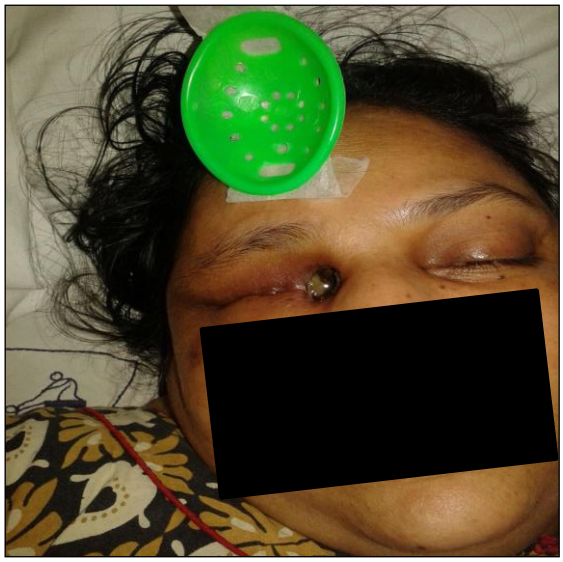

Figure 4: Image after eye exenteration.

Case 3

A 51-year-old female patient with uncontrolled Diabetes mellitus treated in other hospital for diabetic ketoacidosis was admitted in MICU with features of sepsis with shock and multi-organ dysfunction. She was found to have pancytopenia, acute kidney injury, acute hepatitis probably due to ischaemia and hypokalemia. She was immediately started on IV Fluids, vasopressors and antibiotics. Dengue fever, malaria or hepatitis B and C were ruled out with appropriate investigations. She was noticed to have periorbital swelling which was gradually increasing for which ophthalmologist opinion was taken. Her EEG showed focal abnormality suggestive of seizures for which Inj. levipil was started. There was blackish discharge from nose, an ENT opinion was taken and after rhinoscopy, black material from nostril was sent to microbiology for staining. The microbiology report showed rapidly growing mycelial colonies - floccose, dense and hairy appearance, cotton candy growth morphology suggestive of zygomycetes. The fungal preparation with KOH and culture were positive for mucormycosis. The patient was started on lyposomal Inj-amphotericin-B keeping in view the patient having acute kidney injury. CT PNS and MRI neck, larynx, nasopharynx showed diffuse pansinusitis with extension into left retro-orbital fat and pterygoids with no intraparenchymal extension, atrophic chronic optic neuritis and few tiny foci of restriction on diffusion in bilateral cerebellar hemispheres. CT PNS showed mucosal thickening in bilateral maxillary sinuses (left>right) with obliteration of bilateral osteomeatal complex, mucosal thickening in left frontal, sphenoid and ethmoid sinuses and mild deviation of nasal septum to left (Figure 5, 6). An ophthalmology opinion was taken and exenteration of left eye and FESS with surgical debridement of sinuses was done apart from anti-fungal treatment. The sample from sinuses and eyeball were sent to pathology department. She was continued on IV antifungals, vasopressors, ventilatory support and daily hemodialysis. Another anti-fungal Inj. Micafungin was added. The patient continued to be deteriorating and she had cardiac arrest. In this case, the patient had mucormycosis of paranasal sinuses and left orbit with other systemic illness. Even-though FESS for sinuses and left eye exenteration done along with IV Antifungals, she could not survive due to multi-organ failure with acute kidney injury and septic shock.

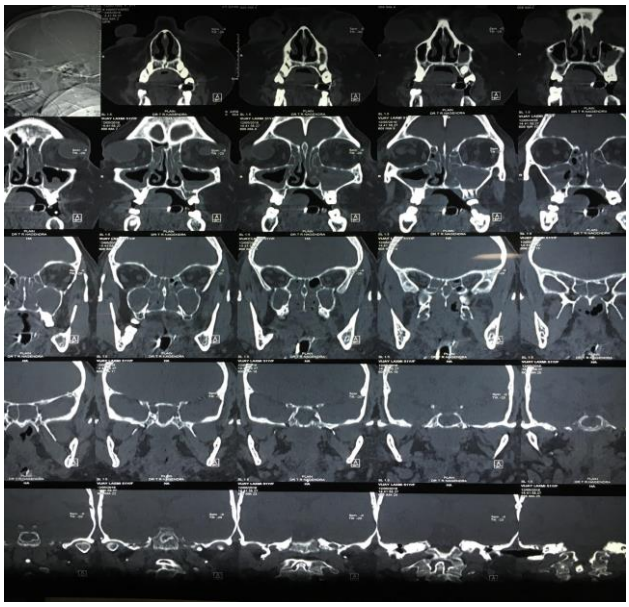

Figure 5: CT PNS mucosal thickening in bilateral maxillary sinuses.

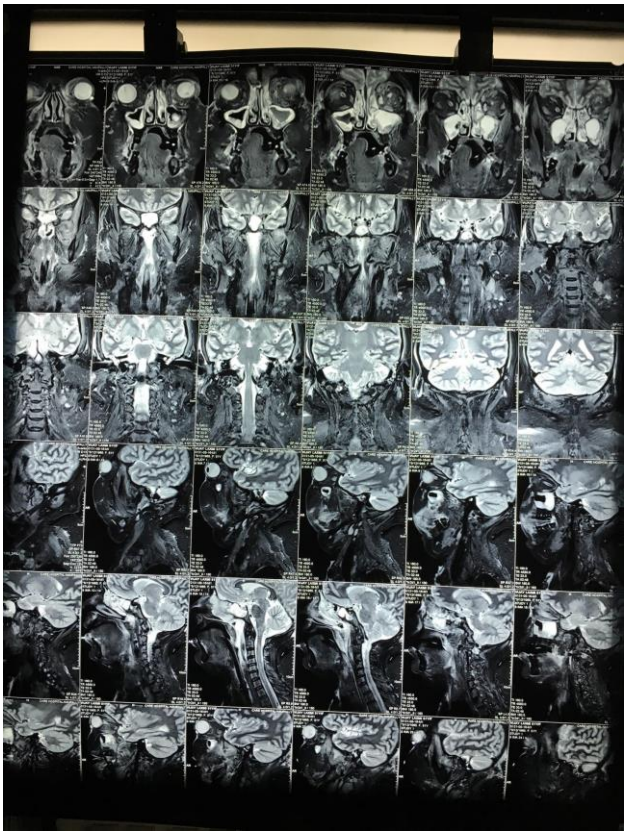

Figure 6: MRI Scan diffuse pansinusitis.

Case 4

A 48-year-old forest officer came with chief complaints of fever with chills for 10 days and yellowish discolouration of eyes for 7 days. Stayed in forest for about 4 days and was fine for one week following which, he developed fever associated with headache and myalgia. There was no history of insect bite. No history of skin itching, bleeding or skin rash. He was a Smoker with 5-6 cigarettes per day and alcoholic with 90ml of brandy/day. No history of DM, HTN, COPD, asthma, TB, CVS Disorders, etc.

On Examination- Patient was conscious, coherent with average built, had Pallor and icterus with no signs of chronic liver disease. He was hemodynamically stable. Abdominal examination showed non-tender distension with no ascites, liver was enlarged with no liver flap.

Investigations showed: Hb. 10.3 g/l, TLC- 14,500/cu.mm, Platelets- 1.14 lakhs/cu.mm, Urea- 289mg/dl, Creatinine- 2.8mg/dl, Serum electrolytes normal except sodium of 125meq/l. Bilirubin- 43.8mg/dl, Direct : 22.9 mg%. SGOT-136IU/L, SGPT-51IU/L, Albumin - 2.8gm/l, INR-1.07

Serum Ammonia - 78, Serum LDH - 1915 IU/L, Serum CPK - 182 IU/L, Serum lactate - 23.8 mg/dl, Procalcitonin- 32.5 ng/ml. Chest X-ray - Normal. ABG - compensated metabolic acidosis. Peripheral smear for malarial parasite revealed gametocytes of P.falciparum and antigen test was positive (Parasite'F').

Viral Markers: HIV, HbsAg and Anti HCV Antibodies - Negative, Dengue Serology negative, Leptospira Serology negative, ANA negative. Urine culture No Growth.

Abdominal US showed: Mild Hepatosplenomegaly.

He was started on I.V Piperacillin + Tazobactam 2.25gm T.I.D, I.V Artesunate 120mg O.D, Oral Doxycycline 100mg O.D, Rifaximin 400mg T.I.D, Oral Lactulose, Liver supplements, Thiamine and INJ. Fondaparinux, One session of bicarbonate hemodialysis for metabolic acidosis. On day nine of admission he had sudden onset of haemetemesis and malaena with significant drop of hemoglobin (8.0 to 5.4g/l). Two units of blood transfusion given, I.V pantaprazole 80mg (stat) followed by 8mg/hr infusion started.

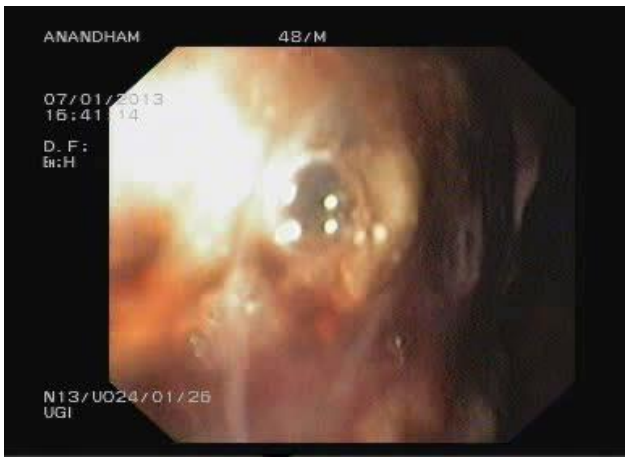

Figure 7: UGI Endoscopy with fresh oozing of blood with old blood clot.

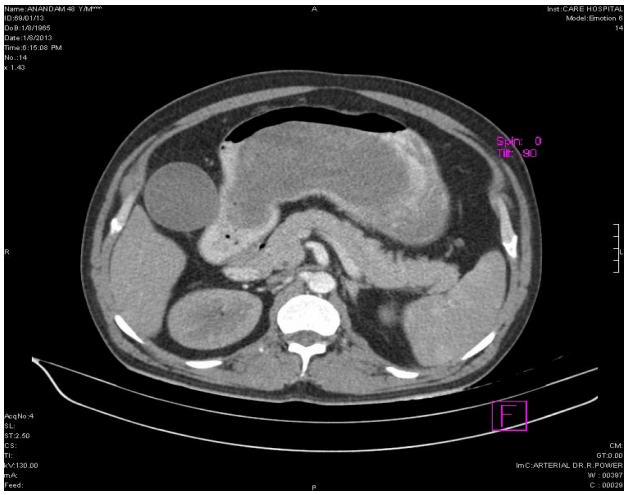

Figure 8: CECT abdomen large clot in gastric lumen, focal thickening of fundal mucosa.

UGI endoscopy showed Ulcerated mass with fresh oozing of blood with old blood clot (Figure 7). Adrenaline injection and Argon Plasma coagulation was done. On day 10 patient had another episode of hematemesis with drop in Hb to 4.0gm%.

CT Abdomen with I.V and oral contrast revealed - large clot in gastric lumen, focal thickening of fundal mucosa (Figure 8), SOL in spleen s/o splenic infarct. Patient underwent emergency laparotomy, Operative findings include ulcer over the greater curvature, adjacent to the posterior wall of stomach - extending up to the fundus and gastroesophageal junction. Lumen of the stomach - filled with blood clots (Figure 9). Gangrenous part of the stomach excised, edges were freshened, sutured, blood clots evacuated. Feeding jejunostomy was done.

The excised edges were sent for HPE showed infiltration by mucor species of blood vessels as well as mucosa of the stomach wall. Fungal culture of the stomach specimen also grew mucor species (Figure 10) after three days. Patient was given I.V Amphotericin B (conventional preparation) at a dose of 0.5mg/kg/day as an infusion in 5% dextrose over six hours in a day. Renal functions and blood counts along with electrolytes were closely monitored. Hypokalaemia was corrected. On the ninth postoperative day, gastrograffin meal imaging was performed, there was no gastric leakage, hence oral feeding commenced. Patient's surgical wound healed well.

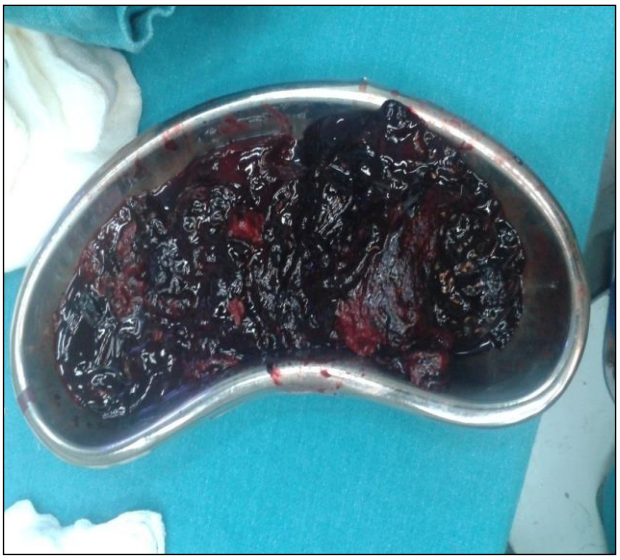

Figure 9: The excised mass with clots.

On the twelfth post-operative day, patient developed recurrence of GI Bleed with drop in Hb. Advised: laparotomy and re-exploration for possible sub-total gastrectomy, the family did not consent. Posaconazole suspension (400mg twice daily) was also added to regimen. Over next few days, patient responded well with no further UGI bleeding. Patient is under follow up and remains clinically well.

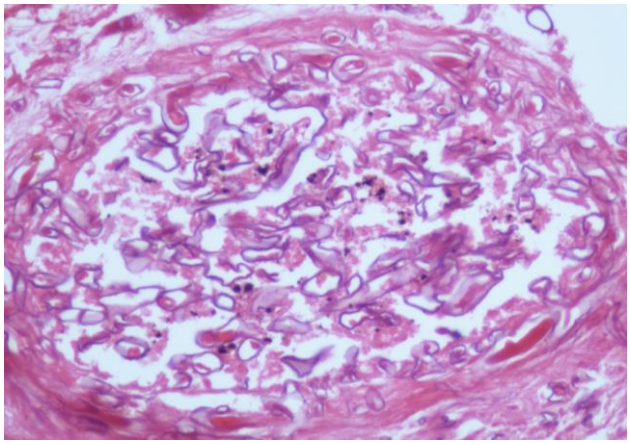

Figure 10: HPE infiltration by mucor species of blood vessels as well as mucosa of the stomach wall.

Case 5

A 60-years-old female operated on emergency basis for DU perforation. She was kept on mechanical ventilation and vasopressors support. Patient was shifted from surgical side to medicine. Subsequently she was weaned off from ventilation and vasopressors. On examination we reported food material coming out from surgical site. We have re-opened the sutures and found food material coming out from body from greater curvature. Biopsy was taken from edges of ulcer and HPE with special stains as sent, which revealed mucormycosis species (Figure 11). Subsequently patient developed septic shock with Multi-organ dysfunction. The patient vitals deteriorated and the patient had a fatal outcome.

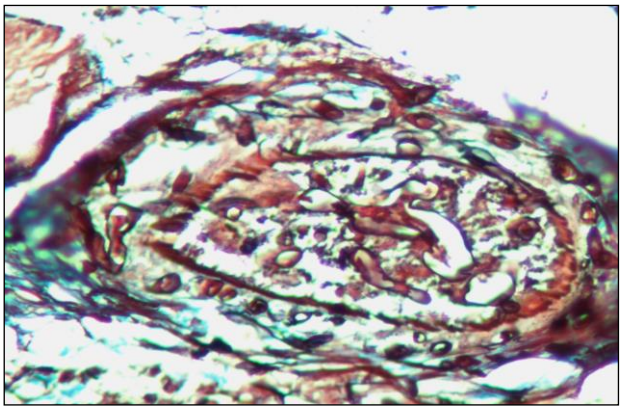

Figure 11: HPE with special stains inflammation and necrotic tissue bit with fungal hyphae with morphological features positive for mucormycosis with zygomycetes species.

Clinical manifestations

As previously mentioned, mucormycosis occurs mainly in those patients with diabetes mellitus and ketoacidosis, in patients with haematological malignancies, especially neutropenia or graft vs. host disease, in solid-organ

transplant patients and in patients receiving high doses of corticosteroids.[3,11,13-36] Treatment with deferoxamine in patients with iron and aluminium overload has been associated with mucormycosis, although the introduction of erythropoietin has significantly decreased the use of deferoxamine, and therefore this factor is becoming progressively less frequent in recent past.[1,13,37-57] Mucormycosis may occur after traumatic inoculation, especially in those cases where the inoculation is accompanied by contamination with water and soil. Mucormycosis manifests most commonly in the sinuses (39%) followed by lungs (24%), skin (19%), brain (9%), and gastrointestinal tract (7%), in the form of disseminated disease (6%), and in other sites (6%).[58] With the exception of rhino-cerebral and cutaneous mucormycosis, the clinical diagnosis of mucormycosis is difficult, and is often made at a late stage of the disease or post-mortem.[59]

Rhino-cerebral mucormycosis

Rhino-cerebral mucormycosis, which should be termed rhino-sinus mucormycosis, accounts upto 33% to 50% of all cases of mucormycosis. Clinical manifestations may start with necrosis of the palate or sinuses, which may progress towards the orbit before reaching intra-cranial structures.[60,61] The most frequent symptoms include fever, obnubilation, amaurosis, proptosis, epistaxis, facial paralysis and signs of invasion of the trigeminal nerve. Thrombosis of the cavernous sinuses and cranial invasion may be consequences of unresolved rhino-sinus mucormycosis. Black sores on the palate or nasal mucosa are very suggestive of mucormycosis in the appropriate clinical context, although they may not be present in 50% of cases.[62,63] The rate of mortality of rhino-orbito-cerebral mucormycosis is still very high and ranges from 30% to 69%.[1,64,65] Indicators of poor prognosis include a delay in treatment of more than 6 days, evidence of intra-cranial invasion, bilateral involvement, invasion of the palate, and the presence of haematological malignancies.[65,66]

In this case series, one patient had rhinoorbitocerebral mucormycosis which responded well to FESS, eye exenteration and irrigation of cavity with amphotericin-B along with systemic antifungals, whereas other with same had fatal outcome.

Respiratory manifestations

Invasion of the lung is the second most common clinical manifestation and follows the inhalation of spores. Manifestations are non-specific and include fever, haemoptysis and pleural pain. Symptoms may appear after near-drowning episodes, and the differential diagnosis of necrotising pneumonia or lung abscesses should be considered.[67,68] The radiological presentation of mucormycosis is similar to that of invasive aspergillosis, and both tend to show vascular invasion and thrombosis, followed by tissue necrosis. Presentation includes cuneiform pulmonary infiltrates, pulmonary nodules and cavitated lesions, including the 'halo' sign.[69,70] The rate of mortality associated with pulmonary mucormycosis is high, and may be over 60%.

Central nervous system involvement

Mucormycosis of the central nervous system may be part of the progression of the disease from a rhino-orbital route or a clinical manifestation limited to the central nervous system.[24,71-74] Delayed diagnosis of CNS involvement may explain the high rate of associated mortality.

Cutaneous involvement

The number of cases of cutaneous and soft tissue mucormycosis has increased during the last few years. This condition can occur on intact skin or following the rupture of barriers, e.g., through surgery, trauma or burns.[75-87] Sometimes, the infection begins at catheter insertion sites or even after insect bites.[9,88-91] It has also been described after the use of contaminated dressings and intramuscular injections.[92,93] In extensive lesions, a cotton-like growth may be observed over the surface of the tissues, a clinical sign known as 'hairy pus'. Rapid diagnosis of cutaneous mucormycosis may explain the lower rate of associated mortality.

Other clinical manifestations

Mucormycosis can involve any part of the digestive tract, with a mortality rate of 98% according to a published series of 25 accumulated cases.[32,94,95-113] Mucormycosis may be the cause of endocarditis in natural or prosthetic valves and has been reported to cause invasion and obstruction of the great vessels.[114-118] The clinical manifestations are extremely rare and the rate of mortality is very high.[31,118-120] The agents responsible for mucormycosis can affect the kidneys and, in these cases, nephrectomy is an essential part of treatment.[29,121-125] Osteomyelitis is another rare form of mucormycosis, and bone lesions are usually adjacent to other forms of mucormycosis . It has been described at the base of the cranium, in the bones of the feet and hands and in the humerus, tibia, femur and vertebrae. Mucormycosis is an extremely rare form of joint infection.[126-134]

Some cases of peritoneal mucormycosis have been described in patients undergoing continuous ambulatory peritoneal dialysis. These are usually complications of previous bacterial peritonitis.[135-139]

In this case study, authors found two cases of mucormycosis inhabiting Gastrointestinal tract. One patient responded well and other had fatal outcome.

Diagnosis

The diagnosis of mucormycosis relies upon the identification of organisms in tissue by histopathology

with culture confirmation. However, culture often yields no growth, and histopathologic identification of an organism with a structure typical of Mucorales may provide the only evidence of infection. A clinician must think of this entity in the appropriate clinical setting and pursue invasive testing in order to establish a diagnosis as early as possible. On the other hand, the agents of mucormycosis can colonize the airways or be contaminants in cultures, and the isolation of these fungi in a culture does not necessarily prove infection. Interpreting the culture results in the context of the patient's signs and symptoms and underlying disease are necessary to determine whether antifungal therapy should be given.

Serum tests, such as the 1,3-beta-D-glucan assay and the Aspergillus galactomannan assay, are being used with increased frequency in patients suspected of having an invasive fungal infection. The agents of mucormycosis do not share these cell wall components and neither test is positive in patients with mucormycosis. (See Diagnosis of invasive aspergillosis, section on Beta-D-glucan assay and Diagnosis of invasive aspergillosis, section on 'Galactomannan antigen detection'.)

Investigational studies have demonstrated the feasibility of using polymerase chain reaction (PCR)-based techniques on histologic specimens.[140-142] In one study of patients with proven mucormycosis, among 12 cases that were positive by culture, 10 were also positive by PCR and sequencing was concordant with culture results to the genus level in 9.[141] Among 15 culture-negative cases, PCR was positive and sequencing allowed genus identification in 12. The PCR-based technique used in this study appears promising for establishing the diagnosis of mucormycosis when cultures are negative.

In addition to traditional culture techniques and PCR with sequencing, matrix-assisted laser desorption ionization-time of flight (M ALDI-TOF) mass spectrometry can be used to identify the causative species from culture specimens.[143-145]

Rhino-orbital-cerebral infection- The presence of mucormycosis should be suspected in high-risk patients, especially those who have diabetes and metabolic acidosis and who present with sinusitis, altered mentation, and/or infarcted tissue in the nose or palate.

Endoscopic evaluation of the sinuses should be performed to look for tissue necrosis and to obtain specimens.

Further evaluation includes imaging of the head with either computed tomography (CT) or magnetic resonance imaging (MRI) to gauge sinus involvement and to evaluate contiguous structures such as the eyes and brain.[146] In a study of 23 immunocompromised patients with fungal sinusitis, CT findings included severe soft tissue edema of the nasal cavity mucosa (turbinates, lateral nasal wall and floor, and septum) in 21 patients, sinus mucoperiosteal thickening in 21 patients, bone erosion in 8 patients, orbital invasion in 6 patients, facial soft tissue swelling in 5 patients, and retroantral fat pad thickening in 2 patients.[147]

Pulmonary infection- The diagnosis of pulmonary mucormycosis is difficult. Chest radiographs or CT scans may demonstrate focal consolidation, masses, pleural effusions, or multiple nodules.[42] A halo sign (ground glass attenuation surrounding a nodule) is characteristic of angioinvasive fungi, but a reversed halo sign, a focal area of ground glass attenuation surrounded by a ring of consolidation, has also been reported. Mucormycosis appears to be the most common condition to cause the reversed halo sign in immunocompromised hosts. In a retrospective study that included 189 patients with proven or probable fungal pneumonia, the reversed halo sign was seen in 7 of 37 patients with mucormycosis (19 percent), 1 of 132 patients with invasive aspergillosis (<1 percent), and none of 20 patients with fusariosis. Radiographic evidence of infarction with cavitary lesions and an air crescent sign is unusual.[68] In a series of 45 cases, the following radiographic features were independent predictors of mucormycosis and helped to differentiate it from aspergillosis: concomitant sinusitis, >10 pulmonary nodules on CT scan, pleural effusion, and prior voriconazole prophylaxis.

Sputum or bronchoalveolar lavage (BAL) specimens can show the char acteristic broad nonseptate hyphae, which is often the first indicator of mucormycosis.[43] However, in one case series, only 25 percent of sputum or BAL specimens were positive premortem. Hyphae can also be demonstrated on lung biopsy.

Histopathological testing does not provide the genus and species, and should therefore be complemented with culture. Histological invasion, particularly of vessels, by wide, non-septate hyphae branched at right angles is diagnostic in an appropriate clinical context.

Treatment

Optimal therapy of mucormycosis involves reversal of predisposing conditions (if possible), surgical debridement, and prompt antifungal therapy. A prolonged course of a lipid preparation of intravenous amphotericin B (5 mg/kg with higher doses possibly given for CNS disease) should be started early. Based on in vitro susceptibility, posaconazole tablets 300 mg/ day orally is generally used after disease has been stabilized. Combination therapy with amphotericin and posaconazole is not proven but is commonly used because of the poor response to monotherapy. Isavuconazole has in vitro activity against some agents of mucormycosis. Other azoles are not effective. There are limited data suggesting beneficial synergistic activity when amphotericin and caspofungin are used in combination for mucormycosis. Despite favorable animal

studies, a pilot study in humans incorporating adjunctive iron chelation therapy with deferasirox demonstrated a higher mortality rate than antifungal therapy alone. Control of diabetes and other underlying conditions, along with extensive repeated surgical removal of necrotic, nonperfused tissue, is essential. Even when these measures are introduced in a timely fashion, the prognosis remains guarded.

Outcome

The overall rate of mortality of mucormycosis is approximately 40%, but this rate depends on the clinical presentation of the disease, the underlying disease, surgery, and the extent of the infection.[8,64,60] Survival rates vary according to the focus of the infection: cutaneous isolated, 90%; sinusitis without cerebral involvement, 87%; rhino-cerebral manifestation, 45%; pulmonary forms, 36%; focal cerebral manifestation, 33%; disseminated disease, 16%; and gastrointestinal involvement, 10%.

CONCLUSION

Authors describe case series of five mucormycosis all of which are biopsy proven. Four out of them are diabetic and one among them is in diabetic keto acidosis. Diagnosing mucormycosis may be difficult and is often delayed because of varied clinical presentation. While three patients had involvement of para nasal sinuses, one patient presented with sepsis and subsequently found to have mucormycosis of stomach. Another patient presented with perforation of duodenal ulcer. All five patients were managed in a multidisciplinary approach with emphasis on prompt anti-fungal therapy, surgical debridement (in form of FESS, exenteration of eye ball, resection of involved tissue etc.,) and addressing risk factors such as control of blood sugars and correction of ketosis. Authors conclude that treating clinicians need to have very high index of suspicion to detect this infection in early phase of illness, good coordination with microbiologists and pathologists to establish diagnosis, so that treatment can be initiated soon for better outcome.

Funding: No funding sources
Conflict of interest: None declared
Ethical approval: Not required

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Cite this article as: Jaju MR, Sagar RV, Srivastav SK, Singh R, Kumar KV, Faisal M. Different types of mucormycosis: case series. Int J Res Med Sci 2020;8:2284-96.