Section A-Research paper

Mucormycosis a dreadful coinfection with Covid 19.

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Abstract:
Covid 19, the global pandemic of the century in its second wave had shown its bad face with another dreaded infection. Initially, treating Covid 19 itself was a challenge to the physicians. After the autopsy study by Laura Falasca et al in Italy it threw light on the management which is accepted and followed till date globally. In spite of handling the infection effectively, the immunocompromised state or diabetic comorbidity in these covid patients had invited the fungal infection mucormycosis. This happens in otherwise healthy patients who after getting infected with covid become immunocompromised. A long stay in intensive care unit, use of ventilator or steroid therapy is considered the possible cause of the disease. Both the infections have few parallel relation; the route of entry nasal or oral and the pathogenesis which is angioinvasion followed by thrombus formation and ischemia. Identifying the early signs of mucormycosis providing appropriate management is as essential as identifying and treating covid itself. This article is focused on the etiopathogenesis, clinical and laboratory diagnosis and management of Mucormycosis. Any delay in treatment of mucormycosis would cause permanent disfigurement to the patient or may be fatal too.

Keywords:
Mucormycosis, covid 19, coinfection, immunocompromised, antifungal therapy.
Running Title: Covid19 &Mucormycosis

Introduction:

The global pandemic of coronavirus 2019 (Covid 19), infected around 161,177,376 people with a mortality of 3,364,178 as of May 2021 with around 6 lakh to 7 lakh cases reported every 24 hours globally.[1] This disease leads to acute respiratory distress syndrome thereby giving opportunity for other respiratory co-infection to occur. Mucormycosis is one such fatal co-infection occurring in patients with Covid 19.[2]Mucormycosis, also known as phycomycosis is an acute opportunistic infection caused by a saprophytic fungus most seen in soil, bread moulds, decaying fruits and vegetables. The mycotic organisms belong to the order of mucorales of the class of phycomycetes; although other genera are associated with this disease like mucor and rhizopus.[3] The first case of this fungal infection was described by Paltauf in 1885 and later the disease got little recognition until 1942 when Gregory reported cases of fatal central nervous system mucormycosis.[4] After that, many other cases have been reported in the literature so far. It may occur as a post covid infection also. Usually mucormycosis is common with immunocompromised patients; its coexistence with Covid 19 is still not vivid. Mucormycosis had expressed itself in Covid patients without any comorbidity or immunocompromised state but who had been in intensive care unit, ventilators and steroid therapy which could be attributed to the cause of this opportunistic infection[5].

Etiopathogenesis:

Naturally, the portal of entry of the coronavirus is the respiratory route (either nose or mouth). They harbor in the throat and nose of the host, also they harbor themselves in the paranasal air sinuses.The virus attaches itself to the host cell, penetrates the cell by endocytosis, the viral RNA enters the nucleus, replicates; biosynthesis of viral particles with the viral mRNA occurs followed by maturation and release.[6] The virus has a spike (S), membrane (M), envelope(E), nucleocapsid (N). The spike has glycoprotein's which determines its diversity.[7] The spike is responsible for adherence and fusion of the virus to the host cell and angiotensin converting enzymes serve to be structural and functional receptors. These receptors are prevalent in lungs, heart, ileum, kidney and bladder.[8] This attributes to the site specific clinical symptoms in patients with Covid. The host responds by its immune system to destroy the cells. Meanwhile there is an increase in inflammatory products like cytokines. The structure similar to viral spike protein is present in the blood vessels; hence the immune system recognizes them also as a foreign protein and acts on them damaging the endothelial cells of the blood vessels resulting in accumulation of blood cells to that site leading to clot formation. This clot blocks the passage of blood flow leading to ischemia and hypoxia results.

Mucormycosis, being an infection of immunocompromised hosts, takes the opportunity of the compromised state in the Covid patients. The spores of the fungi can be ingested or inhaled from duct or air and oral mucormycosis can also be from direct contamination of open oral wounds.[9] Morphologic differences in sexual and asexual pores as well as in other features serve to differentiate the mucorales into various genera.In Rhizopus, the sporangiophores arise from the node of the stolons directly opposite the rhizoides.In Mucor, no rhizoides are present at

the point of attachment of the sporangiophores to the stolons.[10]Rhizopus accounts for 90% of the cases involving head and neck region. Microscopically, mucormycosis appears as a broad, non septate hyphae that exhibit obtuse or right angle branching.[3,9] The sites most commonly involved by mucormycosis infections are the lungs, brain and gastrointestinal tract. Lesions can also occur on skin and oral mucosa. However, occasional cases arise in the central nervous system or cardiovascular system or follow surgical procedures or severe burns.[11] The mucorales have been cultured from the oral cavity, nasal passage and pharynx of healthy individuals without any clinical signs of infection. Invariably, this disease manifests when the organisms affect an immunocompromised patient with severe poorly controlled diabetic ketoacidosis, or in patients with hematological malignant conditions or other debilitating systemic conditions. In diabetes mellitus, acidosis compromises the phagocytic ability of white blood cells thereby affecting the host immunity.[12] Healthy individuals are rarely affected by the disease and those affected have predisposing risk factors such as history of tooth extraction, pneumonia, severe burns, gastrointestinal and rhinocerebral infections.[13] However, prolonged or high dose steroid therapy can bring down the immune status of the patient who is otherwise healthy. This is one possible attribute towards the occurrence of mucormycosis in covid 19 patients.

Looking into the pathogenesis, this too is an angioinvasive infection which has thrombosis and necrosis as hallmark signs. They fungal ligand do adhere to the endothelium, destroy it and cause occlusion too. There are theories supporting mycotoxins too with mucormycosis. [14] Both sharing a similar pathway of cell destruction causes coinfection. Poor oral hygiene, intensive care unit exposure, ventilators, compromised immune status along with any comorbidity contributes to the infection.

Clinical Features in Covid Associated Mucormycosis:

The clinical presentation of mucormycosis depends upon the site of entry of microorganisms and the organ systems involved. Frequency of the disease depends upon the prevalence of different high risk populations and it is difficult to report its prevalence. According to a review, males were more commonly affected than females and also had a mean age of 52.21 years as the age of patients affected with the disease.[9]

The most common site in the maxillofacial region includes rhinocerebral which involves the oral cavity, maxilla, palate, nose, paranasal sinuses, orbit and central nervous system. Other forms are cutaneous, gastrointestinal, pulmonary and disseminated.[3]Oral mucormycosis is mainly caused by inhalation of spores or direct contamination of open oral wounds. Angioinvasion of mucorales and its spores into the blood vessels lead to the formation of thrombus causing progressive necrosis of associated hard and soft tissues. This necrotic tissue is a nidus for organism growth and it thrives there and subsequently invades the surrounding tissue through blood vessels.[14] Early symptoms include facial cellulitis, periorbital edema and nasal inflammation followed by widespread tissue necrosis. Also the patients present with acute sinusitis with fever, nasal congestion, purulent nasal discharge, headache and facial pain.

Rhino-orbital mucormycosis is the most common type of mucormycosis occurring in Covid patients. List of signs and symptoms that should be considered to be "red flags" includes cranial nerve palsy, diplopia, sinus pain, proptosis, periorbital swelling, orbital apex syndrome and ulcers of the palate.[15]Mritika Sen et al reported six cases of mucormycosis with the above features and all were type 2 diabetic patients which establish their compromised immune status. Zesemayat K. Mekonnen et al also had mentioned in his report a patient with diabetes and acute respiratory distress ending in a fatal mucormycosis. Sandeep Sarkar et al had reported 10 cases of mucormycosis in post covid patients who had taken dexamethasone during their treatment and 4 of them had diabetic ketoacidosis. Marina Sandalha reported a case of a young female (32 Years) with orbital mucormycosis post covid management.[16]. S Sharma et al in his prospective study in 2020 reported twenty-three patients presented with mucormycosis, all had an association with coronavirus disease 2019. The ethmoids (100 percent) were the most common sinuses affected. Intra-orbital extension was seen in 43.47 per cent of cases, while intracranial extension was only seen in 8.69 per cent. Diabetes mellitus was present in 21 of 23 cases, and was uncontrolled in 12 cases. All patients had a history of steroid use during their coronavirus treatment. All the reports stated diabetes or steroid therapy as a possible cause for the opportunistic fungi to grow.[17]

Investigation:

The important prerequisites for the diagnosis of mucormycosis are a high index of suspicion, recognition of host factors and prompt assessment of clinical manifestations. Radiologically, thoracic CT scans indicate the presence of mucormycosis by presence of reverse halo sign which is a strong indicator of pulmonary mucormycosis. MRI is also used to study the extent of necrosis, paranasal sinus involvement, orbital and cerebral spread. Other emerging imaging techniques are positron emission tomography-computed tomography(PET CT)with 18F-fluorodeoxyglucose (FDG). Endobronchial ultrasound guided fine needle aspiration is also a useful diagnostic tool.[18].

Microscopy and culture of various clinical specimens are the cornerstones of diagnosing mucormycosis. Direct microscopy of the clinical specimens helps in rapid presumptive diagnosis of mucormycosis. Fungal elements may easily be seen on hematoxylin-eosin section, periodic acid-schiff of Grocott-Gomori'smethenamine silver staining which is used to highlight fungal hyphae and also to evaluate morphology.[19]

Histopathology reveals inflammation which may be neutrophilic or granulomatous and also disease is characterized by prominent infarcts and angioinvasion. All mucorales grow rapidly on most fungal culture media such as Sabouraud's agar and potato dextrose agar incubated at 25 C to 30 C. A specific mouse monoclonal Rhizome mucor antibody has been employed for immunohistochemical analysis and also use of in-situ hybridization targeting 5S and 18S ribosomal RNA sequences remains investigational.[20]

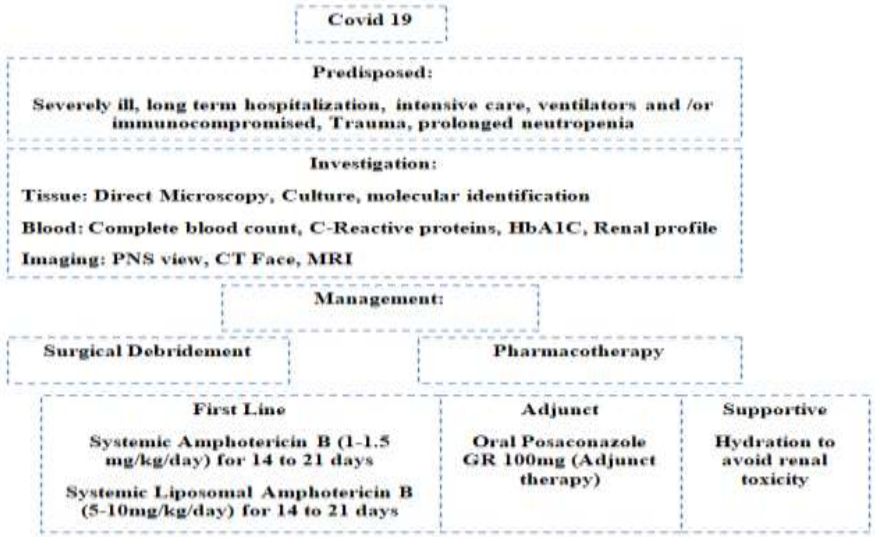
Also, enzyme linked immunosorbent assays, immunoblots and immunodiffusion tests have been tried. Molecular based assays include conventional polymerase chain reaction,

restriction fragment length polymorphism analyses, DNA sequencing of defined gene regions and melt curve analysis of PCR products. Presently, molecular based diagnostic assays can be recommended as valuable add-on tools that complement conventional diagnostic procedures.[21] Differential diagnosis is very important as this disease can have over lapping symptoms and involves similar sites. The various other differential diagnosis includes bacterial sinusitis and allergic fungal sinusitis,aspergillosis, nasal and paranasal malignancies, proptosis, brain tumor, cavernous sinus thrombosis, migraine headache which can have overlapping symptoms to rhinocerebralmucormycosis.[22]

Management:

Management of mucormycosis needs a multidisciplinary approach. A team of oral physician, diabetologist, ophthalmologist and a neuro physician need to treat the disease simultaneously. Hyperglycaemia, diabetic ketoacidosis and metabolic disturbances need to be brought under control. An ophthalmic opinion needs to be sought. European Confederation of Medical Mycology ECMM and Mycosis Study Group Education and Research Consortium (MSGERC) suggest early surgical debridement along with systemic antifungal therapy.[23]Amphotericin B lipid complex, liposomal Amphotericin B and Posaconazole oral suspension are treated as first-line antifungals monotherapy and isavuconazole is supported as salvage therapy.[23][24]Posaconazole is usually combined with liposomal amphotericin B with refractory mycosis or those allergic to amphotericin B. Irrigation of orbit and sinuses with amphotericin B as a local drug delivery enhances the bioavailability of the drug and gives better outcome.[25]Retrobulbar and intraorbital injection of amphotericin B can be considered when aggressive debridement is not possible (1 ml of 3.5 mg/ml with the antecedent retrobulbar injection of anesthetic). [25,26]Hyperbaric oxygen therapy can be administered as the disease has an ischaemic cause. Protocol for mucormycosis management in Covid patients as given by Song et al(Figure 1)[27]

Figure 1: Protocol for management of mucormycosis in Covid Patients



Conclusion:

Mucormycosis is a fatal fungal infection hence it cannot be ignored. If not attended on time it would lead to various complications like have also been reported, like hematoma formation in subarachnoid and subdural regions and intracerebral haemorrhages, ocular complications like ophthalmoplegia, ptosis, proptosis and also conjunctival haemorrhage and edema and orbital cellulitis have also been reported. Meningitis is also a rare but dangerous complication of rhinocerebralmucormycosis. Also, permanent destruction to maxillary and nasal bones can also change the cosmetic facial configuration. Successful management of mucormycosis is based on a multi modal approach including reversal or discontinuation of underlying predisposing factors, early administration of active anti fungal agents at the optimal dose, removal of all infected tissues and use of various adjunctive therapies.

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