INTRODUCTION

Covid-19 or Corona virus disease 2019 is a new disease which is caused due to infection with the novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). It began in Wuhan, China in December 2019, later spreading rapidly throughout the world. World Health Organization declared it to be a pandemic on March 11, 2020. WHO is tracking five variants of concern around the world namely,

Table No:1 Classification of COVID variants

SL.NO	WHO LABEL	PANGO LINEAGE	EARLIEST DOCUMENTED SAMPLES
1	ALPHA	B.1.1.7	United Kingdom
2	BETA	B.1.351	South Africa
3	GAMMA	P.1	Japan
4	DELTA	B.1.617	India
5	OMICRON	B.1.1.529	South Africa

India reported its first case of Covid-19 on 30th January, 2020 in Kerala. As of December 2021, a total of 278,891,403 cases and 5,392,790 deaths were reported in the world and in India reporting 34 765 976 cases and 478 759 deaths due to Covid-19. [5]

In 2020 initiating the second wave. Delta has two mutations linked to increased transmissibility and an ability to escape immune protection making its infection more virulent. Covid -19 usually presents with fever, cough, headache, malaise, breathing difficulties, loss of smell & taste with associated co-infections of bacteria and fungus leading to fulminant pneumonia and death. Patients with Covid-19 disease show high propensity to develop severe opportunistic infections like oropharyngeal Candidiasis, Pneumocystis jiroveci pneumonia, pulmonary aspergillosis, bloodstream candida infections, etc., due to the presence of associated comorbidities like diabetes mellitus, chronic obstructive pulmonary disease and immunocompromised conditions like corticosteroid therapy, ventilation, intensive care unit stay. The Delta variant infection decreased T cells, including CD4 and CD8 cells causing immune dysregulation [45] and led to severe alveolar-interstitial pathology by extensive pulmonary disease [43,44]. This in turn facilitated for the invasive fungal infection of para-nasal sinuses and lung tissue.

Though covid associated Mucormycosis have also been reported in other countries like Iran, Egypt, Mexico and The Netherlands, a massive outbreak was seen in India [7]. The second wave of covid is super-added with the development of Mucormycosis further burdening the overburdened health care system of India. Prior to Covid-19 the prevalence of Mucormycosis in various parts of the world was found to be 34% in Europe, 31% in Asia, 28% in North or South America, 3% in Africa & 3% Australia and New Zealand [2] with overall global incidence of Mucormycosis being 0.005 to 1.7 per million population [1].

Prior to second wave of Covid the prevalence of Mucormycosis in India was found to be 0.14 cases per 1000 population [2][6] or 140 per million population which is 80 times that of developed countries during 2018[6]. Mortality rate was found to be 46% for Rhino-orbito-cerebral form, 76% for pulmonary and 96% for disseminated form in India [1]. the mortality rates in Covid associated Mucormycosis (36%) found to be significantly lower than mortality in non-Covid-19 Mucormycosis [54].

Initially Covid Associated Mucormycosis (CAM) was reported in the western states of Rajasthan, Gujarat and Maharastra with Tamil Nadu witnessing a slow rise of mucor cases in mid may which reached peak by the end of May. Individual states in India declared Mucormycosis a notifiable disease in May 2021[1] in response to the steady rise of CAM and under the directive of Government of India through DO letter dated 19th May and 24th May [8]. There was a total of 51,775[8] cases of Mucormycosis reported all over India till 29th November, 2021. Maharashtra reported highest number of cases in India with 10,366 cases followed by Gujarat reporting 7257 cases, Andhra Pradesh 5107 cases with Tamil Nadu in fourth reporting 5007 cases [8].

JUSTIFICATION

a. Identification of incidence, clinical profile of rhinoorbital Mucormycosis in post covid patients will provide useful knowledge for the prevention, early identification, treatment and reduction of morbidity and mortality.

b. I chose this study due to paucity of studies related to rhino-orbital Mucormycosis in post covid patients in Tamilnadu.

OBJECTIVES

1. To study the incidence of Rhino orbital Mucormycosis in post covid patients
2. To analyse the clinical profile of rhino orbital Mucormycosis in post covid patients
3. To study the risk factors and Immunisation status among the post covid patients developing Mucormycosis
4. Role of mycology, Histopathology and radiology in the diagnosis of Rhino-orbito-cerebral Mucormycosis
5. To develop a diagnostic-criteria for Mucormycosis based on clinical profile and investigations

INTRODUCTION TO MUCORMYCOSIS

Invasive fungal sinusitis is defined as the presence of fungal hyphae within the mucosa, submucosa, bone, or blood vessels on histopathology [9]. Mucormycosis is an opportunistic devastating acute invasive fulminant fungal infection requiring prompt diagnosis and early treatment whose causative agent is ubiquitously present in the environment growing on wet surfaces, dead and decaying vegetable matter.

History:

1855 - first recorded human case of Mucormycosis by Kuchenmeister [2]

1885 - German pathologist Paltauf reported first case of upper airway Mucormycosis - Mycosis Mucorina [3]

1876- Pulmonary Mucormycosis reported in a cancer patient by Fur bringer [2]

1943 - Gregory et al first case of Rhino-orbito-cerebral Mucormycosis [4]

1956 - “Mucormycosis” coined by - American pathologist R.D. Baker [2]

1995 - first known survivor of Mucormycosis [4]

Etiology:

The common etiological fungal agents that cause Mucormycosis belong to:

Kingdom - Fungi

Division - Mucoromycete

Subdivision - Mucoromycotina

Order - Mucorales, and hence the name Mucormycosis

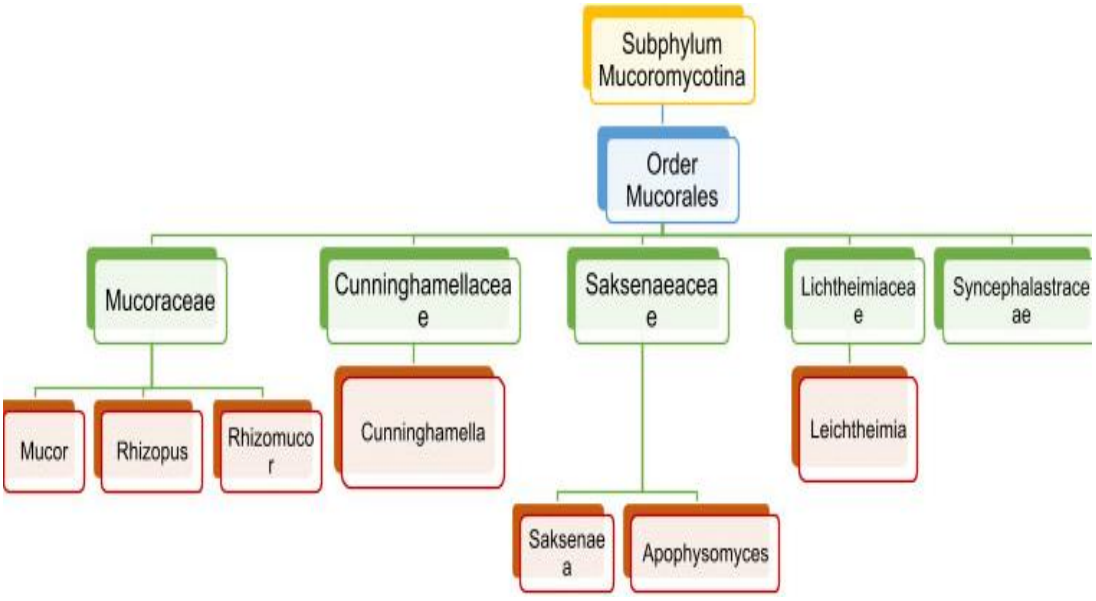
The common genera causing Mucormycosis

A. Rhizopus arrhizus (old - R. oryzae)
B. Rhizopus microspores var, rhizopodiformis
C. Mucor racemosus

} Family Mucoraceae

D. Rhizomucor pusillus
E. Lichtheimia corymbifera (Absidia Species)
F. Apophysomyces Species
G. Cunninghamella bertholletiae – family cunninghamellaceae
H. Syncephalastrum Species
I. Saksenaea Species
J. Cokeromyces recurvatus

Figure no 1- Taxonomy of sub-phylum Mucormycosis [49]



Mucormycosis dubbed as “black fungus” in the Media is a misnomer as the term “black fungus” is used for dematiaceous fungi, which are an entirely different group of fungi. This misnomer may have stemmed from the blackish eschar seen over the face of Mucormycosis patients and has nothing to do with the morphology of the causative fungus.

Classification [10]:

Fungal rhinosinusitis (FRS) is classified as:

Talbot et al; 1980 proposed a classification of invasive fungal sinusitis as

(1) Fulminant aspergillosis,

(2) Rhino cerebral Mucormycosis, and

(3) Aspergilloma.

American Academy of Otolaryngology Head and Neck Surgery and other societies reached a consensus classification in 2009:

Non-Invasive fungal rhinosinusitis:

1) Saprophytic fungal infestation

2) Fungal ball, and

3) Fungus related eosinophilic FRS that includes Allergic Fungal Rhinosinusitis (AFRS)

Invasive fungal rhinosinusitis:

(1) Acute invasive (fulminant) FRS: a fatal rapidly progressive fungal rhinosinusitis evolving within a very short time span. It is Characterized by vascular invasion of fungal hyphae. Mucormycosis is included here.

(2) Chronic invasive FRS, and

(3) Granulomatous invasive FRS [10]

Modes of infection:

Mucormycosis mainly spreads via fungal spore inhalation, food contamination and inoculation of cuts & abrasions over the skin with spores [2] and contaminated medical devices, ventilation systems, disposables in the hospitals including soiled Lenin & bandages with Mucormycosis outbreaks [14].

Cutaneous Mucormycosis spreads via contaminated adhesive bandages, linen, and wooden tongue depressors while rhino-orbital & pulmonary Mucor with contaminated ventilation systems & air conditioners [55].

Groups at high risk of developing Mucormycosis [10] include:

A. Metabolic or systemic disorders:

– Diabetes mellitus

– Haematological disorders, e.g., leukaemia, lymphomas, and aplastic anemia

– Hemochromatosis

– Acquired immunodeficiency syndrome (AIDS)

B. Iatrogenic immunosuppression

– Systemic steroid therapy

– Chemotherapy with neutropenia

– Prolonged antibiotic therapy

Post-organ or stem cell transplantation

C. current and recently treated covid 19 – notifiable epidemic disease

D. Desferrioxamine therapy for iron and aluminium overload (sideroblastic anaemia, beta thalassemia, myelodysplastic syndrome)

Figure no 2 - conditions underlying Mucormycosis [2]:

Types of Mucormycosis:

Various types of Mucormycosis include:

A. Cutaneous

B. Rhino-orbito-cerebral (ROCM)

C. Pulmonary

D. Gastrointestinal

E. Disseminated

Pulmonary & disseminated more common in diabetes and hematological malignancies and disseminated seen among Immunocompromised and burns patients

Mucormycosis is even reported in needle tracks and along the path of polyethylene implants injected for breast implants.

ROCM was due to Rhizopus, pulmonary or disseminated form was caused by Cunninghamella while cutaneous Mucormycosis was linked to Apophysomyces and Saksenaea [15].

Most common infection sites as sinuses amounting to 39%, followed by lungs affected in 24%, with disseminated form in 23% and skin & soft tissue Mucormycosis in 19% [16].

STAGING OF ROCM:

Hanover et al., 2021 proposed a staging system for Rhino-Orbito-Cerebral Mucormycosis [12] as follows -

STAGE I - Involvement of the nasal mucosa

STAGE II - Involvement of paranasal sinuses

STAGE III - Involvement of orbit

STAGE III - Involvement of CNS

Figure no 3 – Staging of ROCM

Staging of Rhino-Orbito-Cerebral Mucormycosis	Symptoms	Signs
Stage 1: Involvement of the nasal mucosa 1a: Limited to the middle turbinate 1b: Involvement of the inferior turbinate or ostium of the nasolacrimal duct 1c: Involvement of the nasal septum 1d: Bilateral nasal mucosal involvement	Nasal stuffiness, nasal discharge, foul smell, epistaxis	Foul-smelling sticky mucoid or black-tinged, or granular or haemorrhagic nasal discharge, nasal mucosal inflammation, erythema, violaceous or blue discoloration, pale ulcer, anesthesia, ischemia, eschar
Stage 2: Involvement of paranasal sinuses 2a: One sinus 2b: Two ipsilateral sinuses 2c: > Two ipsilateral sinuses and/or palate/oral cavity 2d: Bilateral paranasal sinus involvement or involvement of the zygoma or mandible	Symptoms in Stage 1 + facial pain, facial edema, dental pain, systemic symptoms (malaise, fever)	Signs in Stage 1 + unilateral or bilateral, localized or diffuse facial edema, edema localized over the sinuses, localized sinus tenderness
Stage 3: Involvement of the orbit 3a: Nasolacrimal duct, medial orbit, vision unaffected 3b: Diffuse orbital involvement (>1 quadrant or >2 structures), vision unaffected 3c: Central retinal artery or ophthalmic artery occlusion or superior ophthalmic vein thrombosis; involvement of the superior orbital fissure, inferior orbital fissure, orbital apex, loss of vision 3d: Bilateral orbital involvement	Symptoms in Stage 1 and 2 + pain in the eye, proptosis, ptosis, diplopia, loss of vision, infraorbital and facial V1 V2 nerve anesthesia	Signs in Stage 1 and 2 + conjunctival chemoses, isolated ocular motility restriction, ptosis, proptosis, infraorbital nerve anesthesia, central retinal artery occlusion, features of ophthalmic artery occlusion and superior ophthalmic vein thrombosis. V1 and V2 nerve anesthesia, and features of III, IV and VI nerve palsy indicating orbital apex/superior orbital fissure involvement.
Stage 4: Involvement of the CNS 4a: Focal or partial cavernous sinus involvement and/or involvement of the cribriform plate 4b: Diffuse cavernous sinus involvement and/or cavernous sinus thrombosis 4c: Involvement beyond the cavernous sinus, involvement of the skull base, internal carotid artery occlusion, brain infarction 4d: Multifocal or diffuse CNS disease	Symptoms in Stage 1 to 3 + bilateral proptosis, paralysis, altered consciousness, focal seizures	Signs in Stage 1-3 (some features overlap with Stage 3) + V1 and V2 nerve anesthesia, ptosis, and features of III, IV and VI nerve palsy indicate cavernous sinus involvement. Bilaterality of these signs with contralateral orbital edema with no clinico-radiological evidence of paranasal sinus or orbital involvement on the contralateral side indicate cavernous sinus thrombosis. Hemiparesis, altered consciousness and focal seizures indicate brain invasion and infarction.

Clinical Presentations:

Stage I - Symptoms: characterized by Malaise, Low grade fever, Lethargy with persistent Nasal obstruction and discharge, blood stained, mucopurulent or blackish.

Sign: Discharge in the nasal cavity which is foul smelling, mucopurulent, blackish or blood stained, Discoloured nasal mucosa & anaesthesia of the nasal mucosa, Crusting, and ulcer or whitish necrotic slough or blackish eschar over nasal mucosa, middle turbinate, inferior turbinate or septum

Stage II - Symptoms: Presents as Headache, Facial pain, Facial swelling, Dental pain, loosening of teeth, Facial skin redness progressing to blackish discoloration -

Signs: Sinus tenderness, Eschar formation - necrosis of facial skin, Palatal discoloration - bulge & ulcer progressing to - palatal necrosis, eschar & perforation, Gingival swellings, gingival abscess, VII nerve palsy LMN type due to facial Edema

Stage III - Symptoms: has features of Redness & eye pain, Excessive watering, swelling around the eye, Inability to open the eye, Protrusion of eye, Diplopia, Diminished vision progressing to loss of vision

Sign: Chemosis, Ptosis, Proptosis, Restriction of ocular movements ranging from abduction restriction initially progressing to complete restriction of all movements in severe cases, Infraorbital anaesthesia, loss of vision

Stage IV: Symptoms: includes Intractable headache, Altered consciousness, Delirium, Convulsions, Paralysis or Coma

Signs: V1 & V2 anaesthesia, III, IV&VI palsy indicating cavernous sinus thrombosis, Focal seizures, Hemiparesis, Hemiplegia

Pathogenesis:

The hallmark phenotype of Mucormycosis infection is hyphal angioinvasion resulting in thrombosis of blood vessels & infarction and necrosis of tissues [2,20]. Bony invasion causes osteitis and sequestrum formation. In addition, neural and perineural spread is also responsible for spread of the disease in different directions [30]. Brunke et al., 2016 stated that two steps are involved in the host infection with Mucormycosis namely -

1. Surviving within the host by evasion of the immune system, and
2. Host cell damage by perturbing the immune system [21].

A novel host receptor identified as glucose-regulated protein 78 (GRP78) was found to interact specifically and selectively with Mucorales assisting in invasion [22,23] the expression of which is upregulated by high glucose and iron levels [20]. Mucorales damages the epithelial cells by promoting the signalling of platelet-derived growth factor receptor B (PDGFRB) which provided the fungus either necessary growth factors [2]. Gebremariam et al., 2014 found that a protein over the surface of Mucorales spore called Spore Coat (CotH) protein aids in penetration, disruption and damage of host immune cells & mediates endothelial cell invasion by the Mucoralean fungi [24].

Any defects in the hosts immune response, either quantitative (neutropenia) or qualitative (high sugars, ketoacidosis, usage of corticosteroids) facilitates the growth of mucoralean fungal hyphae and invasion.

Corticosteroids acts as a double-edged sword reducing inflammation on one hand while causing reduction of immune activity by reducing White Blood Cell and T-helper cell production paving the way for easy invasion of the fungi. Steroids also cause uncontrolled sugar levels in the blood enabling the growth of Mucor. Cytokine storm, aggravated by high sugars damages the endothelium and the acidotic environment of ketoacidosis facilitates the adherence and invasion of the hyphae.

Various factors enhance the availability of iron to Mucorales like uncontrolled diabetes or ketoacidosis which decreases the affinity of transferrin & ferritin towards iron [31,32]. Mucorales utilizes deferoxamine as a xenosiderophore to pump iron from the host assisting in the hyphal growth [33].

Routes of Spread:

Disease spreads from nasal cavity to maxillary sinus and from the maxillary sinus it spreads Anteriorly to the premaxillary fat & soft tissue,

Posteriorly to the pterygopalatine fossa, superiorly through the roof to involve orbit and Inferior spread causing palatal erosion & necrosis, loosening of teeth.

From the Pterygopalatine fossa, a central hub, Mucor spreads Laterally to the infratemporal fossa resulting in trismus and swelling of the cheek. Inferior spread through the greater palatine foramen and canal leads to palatal erosion and necrosis. Superiorly through the inferior orbital fissure into the orbital apex and then through the superior orbital fissure in to the middle cranial fossa. Posterior spread through palatovaginal canal leads to erosion of pterygoid process and the pterygoid plates and to the greater wing and the middle cranial fossa. Posterior can also spread through vidian canal involving middle cranial fossa and

Perineural spread along the maxillary nerve via foramen rotundum and the mandibular nerve via foramen ovale leads to involvement of the trigeminal ganglion associated with severe pain in its area of supply.

From Ethmoids spreads in to the orbit laterally through the natural foramina and erosion of lamina papyracea, superiorly into fovea, frontal basal meningitis and further intracranial spread. Causes necrosis of middle and superior turbinate Medially.

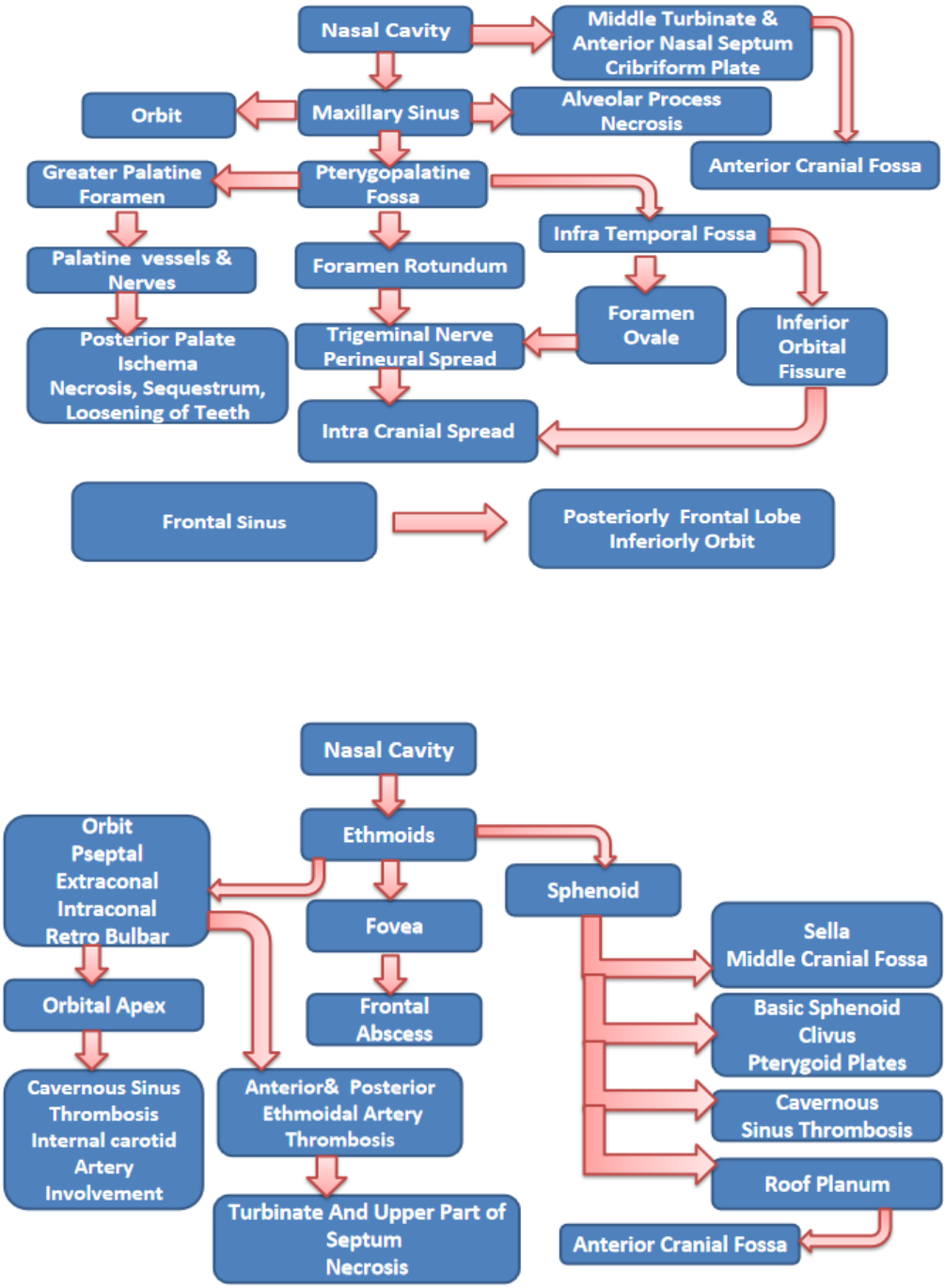
Orbits are involved laterally from the ethmoids and superiorly from the maxillary sinus. Thrombosis of anterior and posterior ethmoid arteries results in necrosis of the turbinates and the upper part of the septum.

Orbital apex involvement leads to optic nerve infarction and further spread causing cavernous sinus thrombosis and internal carotid artery involvement.

From the sphenoid mucor involves the Sella and middle cranial fossa, planum sphenoidale and anterior cranial fossa Superiorly,

Posteriorly involves the clivus, Basi sphenoid and Pterygoid wedge & pterygoid plates. Lateral spread causes cavernous sinus thrombosis.

Figure no 4 – Routes of spread of ROCM



Diagnosis:

Various modes of diagnosis include -

I. Diagnostic Nasal Endoscopy (DNE)

II. Radiology

III. Microbiology

IV. Histopathology

Diagnostic Nasal Endoscopy (DNE):

Findings include blackish or bloodstained nasal discharge, necrotic slough or blackish eschar over nasal mucosa middle turbinate, inferior turbinate or septum

Pathology:

Diagnosis by Histopathology includes three components [10] namely

i. Demonstration of fungal hyphae

ii. Identification of angioinvasion

iii. Identification of bone invasion

Mucoralean fungal hyphae as aseptate or pauci-septate of irregular diameters, thin walled, broad ribbon like with non-dichomatous irregular branching with associated tissue necrosis and angio & bony invasion [3].

The hyphae can be demonstrated using haematoxylins-eosin staining or with special staining techniques like periodic acid-Schiff (PAS) stain and Gomora's (Grocott–

Gomori's) silver methenamine (GMS) preparation that are based on the principle of oxidizing the fungal wall polysaccharides.

Mucorales has a propensity for invading blood vessels [47] in particular the arteries. Features of angioinvasion include dissection of internal elastic lamina away from the media due to hyphal growth, luminal growth of hyphae and endothelial injury with thrombus formation [48]

Microbiology:

Microbiological diagnosis includes

a. Direct brightfield Microscopy
b. Culture
c. Molecular methods
d. Serology

Direct brightfield microscopy:

KOH wet mount examination of nasal discharge and biopsied tissue shows broad (6-16 µm), cenocitic, ribbon like hyphae with irregular wide or right-angled branching [49,50]. fluorescent stains like calcofluor white staining in which the hyphae appear bright green to blue can also be used. Microscopy helps in rapid diagnosis of Mucormycosis [50]. lactophenol cotton blue mount is also widely used.

Culture:

Culture of fungus is difficult and preferably the sample should be obtained before the initiation of antifungals [10]. Commonly used culture media include Sabouraud dextrose or honey agar and Brain & heart infusion agar with antibiotics at an intubation temperature of

28^0C and 35-37^{o}C. Growth can be seen in 24-48 hrs. The colonies grow vigorously, called "lid-lifters," with characteristically white or greyish black, floccose, dense and hairy in appearance. In 50% of cases cultures can be false negative [3,49].

Molecular Methods:

Species identification can be done using newer methods like MALDI-TOF Matrix assisted laser desorption ionization-time of flight mass spectrometry and RT PCR (internal transcribed spacer (ITS) or the 18S, 28s rRNA genes).

Serology:

β-D glucan test and Aspergillus galactomannan assay tests are positive in invasive aspergillus and negative in Mucor mycosis

Radiology:

Common radiological imaging requested are CT paranasal sinuses with orbit - bony and soft tissue windows, MRI paranasal sinuses and MRI Brain.

Middlebrooks et al., observed that in CT usually there is unilateral predominance with involvement of ethmoid, sphenoid and maxillary commonly [52]. Hyperattenuating mucosal thickening, sinus opacification, bony destruction that is subtle or extensive and Fat stranding outside the sinus – intra orbital fat, periorbital fat, nasolacrimal duct, lacrimal sac, pterygopalatine fossa. Extra sinus involvement with intact bony walls hint at the perineural and angio invasive properties of the Mucorales.

Soft tissue involvement and bone marrow infiltration can be discerned well in MRI.

Figure 5 – Common MRI findings

Structure	Abnormalities
Rhinosinus	T2 heterogenous mucosal thickening, turbinate/ mucosal nonenhancement, fluid collections
Extrasinus	Facial subcutaneous soft tissue, pteryopalatine fossa, masticator spaces
Bone	Sinus walls, pterygoid processes, alveolar processes, sinus bordering orbit walls, skull base – destruction, subperiosteal abscesses
Orbital	Preseptal, postseptal inflammatory changes, optic nerve involvement (infarction, perineuritis), apex soft tissue, EOM nonenhancement, Globe
Vascular	Arterial: ICA steno-occlusion, ophthalmic artery occlusion, ECA occlusion, mycotic aneurysms Venous: Cavernous sinus – Direct invasion vs thrombosis/thrombophlebitis, Superior ophthalmic vein thrombosis
CNS	Ischemic changes – sterile infarcts (usually watershed, less commonly territorial), infarcts with secondary fungal invasion Direct invasion – abscesses/granulomas, meningitis, perineural spread via trigeminal nerve, contiguous brainstem involvement

EOM – Extraocular muscle, CNS – Central Nervous System

REVIEW OF LITERATURE

1855 - first recorded human case of Mucormycosis by Kuchenmeister [2]

1877- Pulmonary mucormycosis reported in a cancer patient by Furbringer [2]

1885 - German pathologist Paltauf reported first case of upper airway mucormycosis - Mycosis Mucorina [3]

1939 Foster & Waksman et al., discovered **zinc** to be essential for fungal growth indicating its possibility as a factor for the epidemic

1943 - Gregory et al., first case of rhino-orbito-cerebral Mucormycosis [4]

1955 - first known survivor of Mucormycosis [4]

1956 - "Mucormycosis" coined by - American pathologist R.D. Baker [2]

1967 - Green et al., Addlestone et al.,1975; & Gemba et al.,1986; mainly highlights the bony erosive nature of Mucormycosis

1984 - Ketenci et al., & Yohai et al.,1994; stated that palatal involvement may be seen in later stages of Mucormycosis probably due to direct spread from Maxillary sinus.

1992 - Clary-Meinesz et al., ascertained that exposure to temperatures of 45^0 C and above led to diminished ciliary beat frequency of nasal epithelium with the possibility of even cell death predisposing the nasal mucosa for infections.

1993 - Jones et al., observed palatal ulcer to be the most frequent oral sign in Mucormycosis

1994 - Yohai et al., stated that palatal involvement may be seen in later stages of Mucormycosis probably due to direct spread from Maxillary sinus.

1998 - studies of Silverman & Mancuso., focused on CT changes that appear much earlier in the course of the disease stressing on the invasive nature of Mucormycosis like extra sinus soft tissue involvement in the form of fat stranding especially in the peri antral region

1998 - Kandpal et al., reported perineural spread of the fungal hyphae

2000 - Ribes et al., perceived aggressive growth and invasion of Mucorales spores in patients with increased availability of serum & tissue iron. He opined that palatal necrosis to be caused by extension of Mucor from sinuses

2005; roden et al., ascertained that rhino cerebral form was more frequently associated with diabetes (33%) and then in Haematological malignancies 4%

2005 - Simmons et al., stressed on the fact that hyperbaric oxygen was itself used as an adjunctive therapy to treat mucormycosis

2007 - Aribandi et al., discovered that in certain cases involvement of bone could be insignificant and extra sinus involvement might be appreciated even with intact bony walls of the sinuses and orbit owing to the Angio invasive nature of the Mucoralean fungi [98] recorded the involvement of orbital fat, extraocular muscles, peri antral fat as the harbinger of hyphal invasion. Observed that Leptomeningeal enhancement heralds the Intracranial extension of mucormycosis which can progress to cerebritis, granuloma and cerebral abscess in advanced disease.

2009 - Chakrabarti et al., found diabetes to be the most common underlying risk factor of Mucormycosis in India [19]

2011- Skiada et al., stated that in Europe mucormycosis is most commonly seen in haematological malignancies

Sahasrabudhe et al., identified a total of 14 fungal species in humidifier chambers out of which 4 were virulent

2013 - Jadhav et al., reported 75% colonization rate by fungal species in swabs from respiratory devices including humidifier chambers but none of the fungi were of Mucoralean species [8

Staats et al., discovered **zinc** to be essential for fungal growth indicating its possibility as a factor for the epidemic

Sachdev et al., reported a case series of 6 patients of rhino-orbito-cerebral Mucormycosis with facial palsy.

2014 - Gebremariam et al., found that a protein over the surface of Mucorales spore called Spore Coat (CotH) protein aids in penetration, disruption and damage of host immune cells & mediates endothelial cell invasion by the Mucoralean fungi [24].

Baez and Shiloach et al., noted that **Industrial oxygen** is produced in the same way as medical grade oxygen and high oxygen concentration is proven to hinder the growth of microorganisms

Jain et al., Blackish palatal eschar was noted in Mucormycosis patients

Cornely et al., highlighted the importance of direct microscopy in his study as it allows for rapid and prompt working diagnosis far before culture or histopathology

2015 - Kursun et al., found rhino-orbito-cerebral mucormycosis to be the most common form with diabetes as major risk factor leading to mucormycosis.

Nguyen et al., determined that the prolonged and inappropriate use of higher antibiotics tips the delicate balance between the mycobiome and microbiome of the nasal and respiratory lining promoting infections like mucormycosis

Nicolatou-Galitis et al., found palatal signs to be the presenting complaint along with other signs in 34 cases and found palatal ulcer to be the sole presenting complaint in 8 patients leading to the diagnosis of Mucormycosis [86].

2019 - Jeong et al., in their analysis of case reports over a period of 17 years, stated that rhino cerebral mucormycosis to be commonly associated with diabetes & disseminated form of Mucormycosis to be associated with haematological malignancies and also observed that ROCM was predominantly due to Rhizopus species

Prakash et al., observed underlying haematological malignancies in 6.3% of mucormycosis cases [78].

Singh et al., noticed mucosal discoloration in early stages and blackish eschar involving middle turbinate, septum and inferior turbinate.

2020; Hoang et al., in a study based in USA discerned that chronically elevated blood sugars, that deter the proper functioning of phagocytes and neutrophils, can be expedited by even a short course of steroids predisposing to Mucormycosis [67].

Brandao et al., The various oral signs demonstrated in the study by include palatal discoloration, bulge, ulcer, necrosis and eschar.

2021 - Abdel-Tawab., in a study from Egypt on CT chest severity of Covid-19 identified 47.9% of study population to have less than 50% lung involvement and 23.1% more than 50% lung involvement [94]

Hicks et al., reported 89% [77] and Shankar et al., remarked 86% [76] of the post Covid Mucormycosis patients to be unvaccinated for Covid-19.

Patel et al., found the incidence of CAM to be 65.2% and prevalence to be 0.27% in **hospitalized covid patients** [68].

Muthu et al., reviewed 275 cases of Covid Associated Mucormycosis comparing CAM in India and the rest of the world. They defined CAM as onset of Mucormycosis within 3 months of positive Covid status (<7 early CAM and >7 late CAM). he observed that on an average CAM was reported after approximately 19.5 days from the date of covid-19 positivity with most being late CAM. he accounted 89% of CAM cases to rhino-orbital (ROM) and rhino-orbito-cerebral mucormycosis (ROCM) in India, with the same reported in 64% cases in the rest of the world and other presentations, like pulmonary & disseminated forms, were reported rarely in India. He suggested that disordered immune response in **Covid-19,** inappropriate & excessive use of steroids and antibiotics in the background of diabetes to be the prime guilty factors for CAM epidemic and felt that industrial oxygen has no role in the surge of CAM. He observed **Diabetes mellitus** to be the most frequent underlying risk factor for CAM in India (66.1%) when compared to the rest of the world with (54.8%). Muthu et al., ascertained that the mortality rates in Covid associated

Mucormycosis (36%) to be significantly lower than mortality in non-Covid-19 mucormycosis [54].

A descriptive study by **Mishra et al.,** found 3.36% (32 cases in total) of the patients with covid to develop Mucormycosis. 56.2% of the patients with Mucormycosis had a **history of severe Covid -19.** [76] none of his study population were fully **vaccinated** against Covid while two of them received first dose [76]. 87.5% were diabetics and apart from diabetes, hypertension was the most commonly associated comorbidity in CAM. He found maxilla to be the frequently affected sinus followed by ethmoid and observed orbital involvement in 59.4% cases **radiologically** [76]. he described 90.6% **KOH** positivity, 84.3% showed fungal hyphae in **HPE** and 34.7% had fungus grown on **culture. He concluded that diabetes and inappropriate steroid usage as the contributing factors for the epidemic.**

2020 - El-Kholy et al., in his study in Egypt reported 36 patients with CAM. Mean duration of onset post covid was 17.82 ± 2.97 days with two patients having Mucormycosis along with active covid infection. Mean age was found to be 52.92 ± 11.30 years with pain & numbness over face (75% & 66.7%) and orbital features (63.9%) as the most common clinical features. Diabetes followed by hypertension were the associated comorbidities in the majority. Radiologically ethmoid sinus was most commonly involved followed by sphenoid.

2021 - Sen et al., conducted analysis of 2826 CAM patients reported throughout Indian states. He observed 51.9 to be the mean age of patients developing CAM with 71% being

male[13]. he reported that 28% of the study group were home isolated and 72% required hospitalization for Covid-19 treatment out of which 87% received steroids and 57% needed **oxygen** support for covid-19 management during hospitalization.[13]Sen et al., 2021; concluded that diabetes is a major risk factor for ROCM (78%)[13]Apart from diabetes, hypertension was the most commonly associated comorbidity in CAM and he detected cardiovascular disease in 1.9% and cerebrovascular disease in 0.9% of Post covid patients with Mucormycosis. The study identified that 18% of the CAM patients had no underlying comorbidities apart from Covid -19 and 5% (23) of them received neither O2 nor corticosteroids. Various symptoms of CAM observed by Sen et al., 2021; include orbital or facial pain in 23%, facial or orbital Edema in 21%, vision loss in 19%, proptosis in 11%, ptosis in 11% and nasal stuffiness in 9% [13].. **Signs** of CAM noticed by Sen et al., 2021; were periorbital Edema (33%), loss of vision in (21%), ptosis in (12%) and proptosis in (11%) [13]. he determined the involvement of sinuses in **CT** to be unilateral in 59% and bilateral in 40% [13] with maxilla to be the frequently affected sinus followed by ethmoid's involvement in **MRI** was seen in the form of cavernous sinus thrombosis (53%), internal carotid artery occlusion (18%), temporal lobe abscess (12%), frontal lobe abscess (2.8%) and skull base osteomyelitis (7.1%). 49% of his study population had severity of stage 3b or less and 27% had stage 3c or above. fungal hyphae were detected for 89% in KOH, 19% showed culture positivity and 39% were positive for mucor in histopathology.

Sathish & Chandrika Anton et al.,2021; pointed out that 20.6% of post covid patients had [new] onset diabetes mellitus

2021 - Kumar et al., suspected that industrial oxygen could be a contributing factor for the epidemic of Mucormycosis in Post Covid individuals.

Pal et al., hypothesized that water used in oxygen humidifiers to be the potential reservoir and source of fungal spores

Aranjani et al., & Gupta et al., debated that Mucorales lacks the ability to produce spores in water of humidifiers agitated by the passage of oxygen [80,81]

A study by Hussain et al., showed only 0.89% of CAM patients to be associated with underlying haematological malignancy [6]

2021 - In post covid era Mitra et al., observed palatal involvement in 25% of CAM patients [91] and Sharma et al.,2021; reported blackish eschar over hard palate as oral manifestation in 39% of patients

Mehta et al., identified facial palsy in 17 (8%) out of 196 patients with CAM.

A study by Hussain et al.,2021; showed only 0.89% of CAM patients to be associated with underlying haematological malignancy [6]

Hypodense sinus mucosal thickening, nasal cavities and turbinates were noted on non-contrast CT by Joshi et al., reported maxilla to be the frequently affected sinus followed by ethmoid.

Joshi et al., reported bony erosions in 80% of his study population involving sinus walls, nasal septum, ethmoids, orbital walls, cribriform plate, wings of sphenoid, Sella, skull base and hard palate [104].

Desai et al., found 62% of his study population with bilateral involvement and 44% had predominant right sided and 40% predominant left sided involvement on CT.

Maxilla to be the frequently affected sinus followed by ethmoid and noted that extra sinus soft tissue hypo intensity could be evident even with no bony erosion. Author observed that fungal involvement in CAM to appear as hypo intensity on MRI which can be classically seen as Black Turbinate sign [97]. He reported that Orbital invasion on MRI can be observed in the form of muscle thickening, extra & intraconal fat stranding, optic nerve involvement and orbital apex soft tissue involvement [97]. noted Optic nerve involvement could be discerned as heterogenous enhancement with diffusion restriction on MRI-DWI ADC sequencing [97]. he stated Retro /pre/peri antral soft tissue inflammation could be identified as fat stranding on MRI [97]. Desai et al.,2021; stated early Intracranial extension was discernible in the form of Dural enhancement along Basi frontal or Basi temporal region and in later stages as cerebritis, cavernous sinus & ICA involvement and abscess noted as hypo intensity on T1 & T2 and on contrast had peripheral rim enhancement [97]. Desai et al.,2021; noticed Olfactory bulb involvement on MRI as heterogenous enhancement of the bulb [97].

Sharma et al.,2021 noted ethmoid to be the most common sinus to be involved followed by maxilla

Sekaran et al.,2021 discovered NLR values to be elevated in post Covid Mucormycosis patients.

METHODOLOGY

5.2. STUDY DESIGN:

This study is a hospital based Cross sectional study conducted in RGGGH, MMC, Chennai.

5.3. STUDY POPULATION:

Patient who admitted in Mucor ward in RGGGH, Chennai.

5.4. STUDY DURATION:

MAY 2021 TO SEPTEMBER 2021 (6 months period)

5.5. STUDY AREA:

Rajiv Gandhi Government General Hospital, Chennai - 03.

5.6. INCLUSION CRITERIA:

Post covid patients with new onset rhinoorbital mucormycosis

Those who are willing to give informed consent.

5.7. EXCLUSION CRITERIA:

Patients with pre-existing rhino-orbital mucormycosis

Those who are not willing to give consent

5.8.DATA COLLECTION TOOL:

To collect A detailed clinical history, microbiological and radiological investigations in post covid patients with rhino-orbital mucormycosis

5.9. SAMPLING METHOD:

Convenient sampling

5.10. SAMPLE SIZE CALCULATION:

The sample size was calculated based on the incidence of mucormycosis in Asia[2] was 31 %, with a 95% confidence and an absolute precision of 5%, sample size is derived. Sample size is calculated using the formula:

$N = 3.84 \text{ X } pq / d^2$

N=sample size

Z value for α at 0.05=1.96

p=prevalence=31 %

q=100-p=100-31=69%

d =Absolute precision of 5%

Sample size **$N = 3.84 \text{ X } pq / d^2$**

3.84X31X69 / 5X5 = 328

Considering 10% non-response rate, calculated sample size, **N= 361**

The incidence of covid associated mucormycosis cannot be calculated due to ongoing pandemic, hence we opted for large number of sample size

5.11. ETHICAL CLEARANCE

- Obtained from the institutional ethical committee of Madras Medical College
- Official permission to conduct the study obtained from DEAN , MMC, Chennai

5.12. DATA COLLECTION:

- All hospitalized mucor patient were interviewed using pre tested semi structured questionnaire after obtaining their consent
- We took clinical History, covid related history, Vaccination status, test conducted for fungi, CT, MRI and clinical investigation

5.13. DATA ANALYSIS:

- The data was entered in MS Excel sheet and analysed using SPSS Version 16. Appropriate descriptive and inferential statistics were used to analyse the data.

RESULTS

Figure no 6 - Distribution of Age among study population

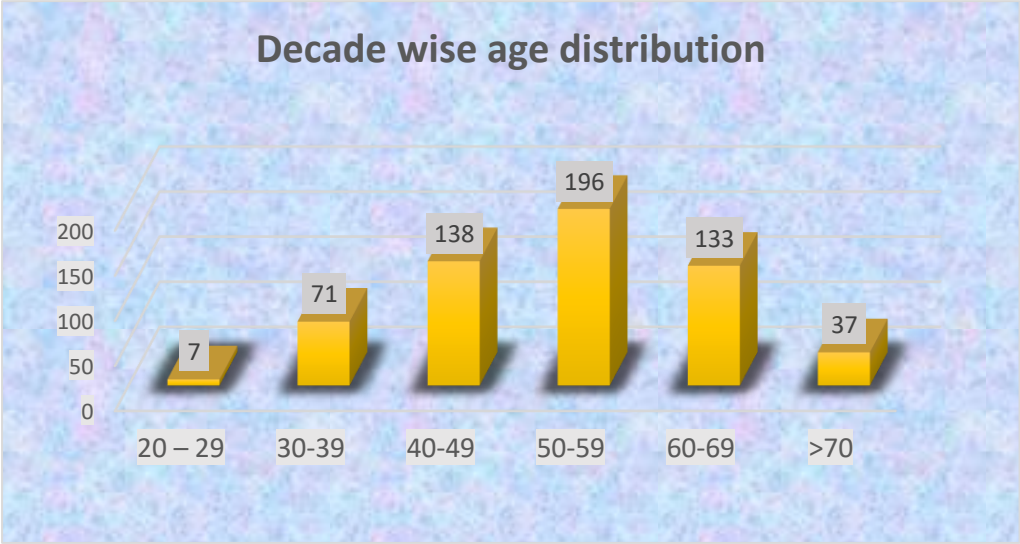


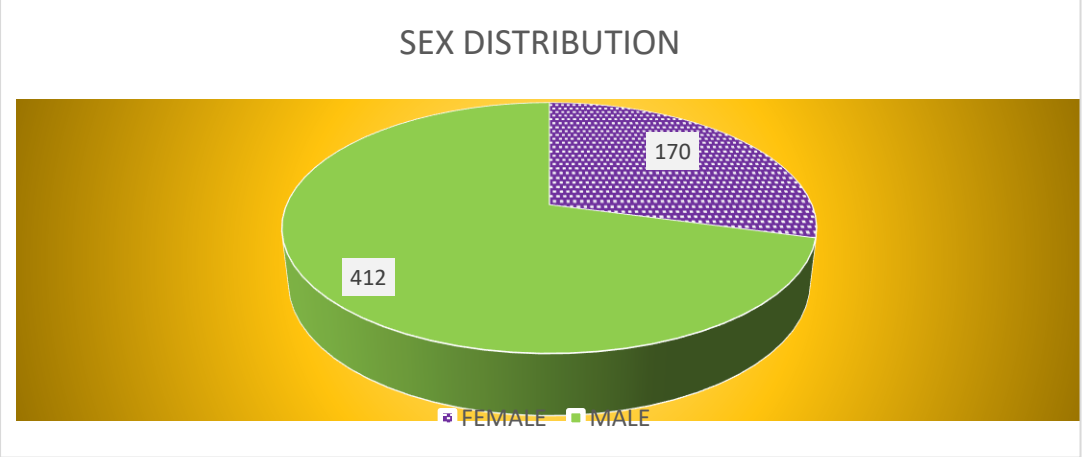
Table no 2 Mean age distribution among Study population

MEAN AGE	SD	MINIMUM	MAXIMUM
52.69	**52.69±10.98**	23	81

In our study population of 582 cases, mean age of developing CAM was found to be 52.57 ± 10.9 years with 1.2% of cases seen in second decade, 12.2% in third decade, 23.7%

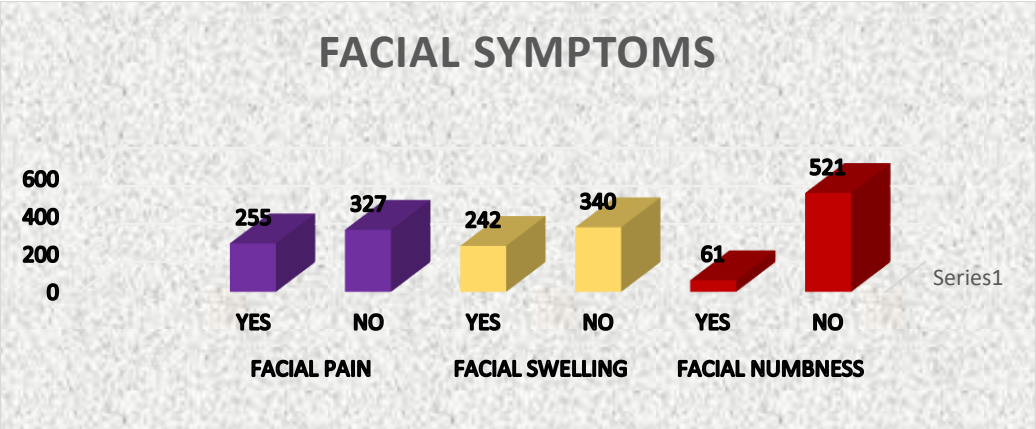
in fourth decade, 33.3% in fifth decade, 22.9% in sixth decade and 6.4% cases seen in seventh decade and above.

Figure no 7 -sex wise distribution of study population



Among 582 study population 70.8% were males and 29.2% were females.

Figure no 8- distribution of facial symptoms among the study population



Among the study population of 582 cases, 255 (43.8%) presented with complaints of facial pain, 242 (41.6%) presented with facial swelling and 61 (10.5%) participants had complaints of facial numbness

Table no 3– distribution of nasal symptoms among study population

NASAL SYMPTOMS		FREQUENCY	PERCENT
NASAL BLOCK	YES	488	83.8
	NO	94	16.2
MUCOID DISCHARGE	YES	385	66.2
	NO	197	33.8
BLOODY DISCHARGE	YES	47	8.1
	NO	535	91.9

Among the study population of 582 cases, 488 (83.8%) presented with complaints of Nasal block, 385 (66.2%) presented with mucoid discharge and 47 (8.1%) participants had complaints of blood-stained nasal discharge

Table no 4- distribution of eye symptoms among study population

OPHTHAL SYMPTOMS		FREQUENCY	PERCENT
EYE PAIN	YES	**181**	31.1
	NO	401	68.9
EYE SWELLING	YES	**153**	26.3
	NO	429	73.7

VISUAL DISTURBANCE	YES	**121**	20.8
	NO	461	79.2

Out of 582 study population 181 (31.1%) among the study population of 582 presented with eye pain.153 (26.3%) had a history of eye swelling and the visual disturbance was a presenting complaint in 121 cases (20.8%)

Table no 5- distribution of headache among study population

HEADACHE	FREQUENCY	PERCENT
YES	**264**	45.4
NO	318	54.6

264 (45.4%) cases among the study population of 582 had a history of headache

Table No:6 Facial Palsy among study participants

FACIAL PALSY	FREQUENCY	PERCENT
YES	**23**	4.0
NO	559	96.0

Out of the total 582 study population 23 (4%) population presented with lower motor neuron type of facial palsy.

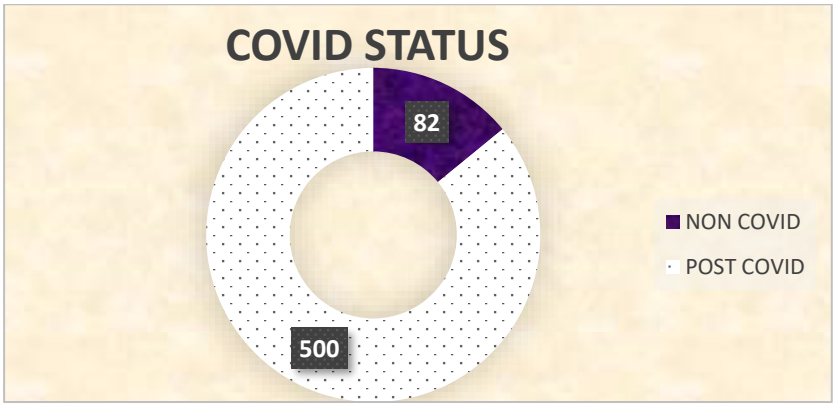
Table No: 7 Distribution of Dental symptoms among study participants

VARIABLES		FREQUENCY	PERCENT
TOOTHACHE	YES	**84**	14.4
	NO	498	85.6

TOOTH LOOSENING	YES	**20**	3.5
	NO	562	96.5

84 (14.4%) cases out of the 582-study population presented with toothache as a complaint. Loosening of tooth was a complaint among 20 (3.5%) of the study population

Figure no 9 - distribution of Covid status among study population



Out of the 582-study population 500 (85.9%) had a history of Covid-19 infection and 82 (14.1%) were Non covid.

Table no 8 - distribution of covid CT severity scale among study population

COVID SEVERITY	FREQUENCY	PERCENT
GRADE 1	111	19.1
GRADE 2	**145**	30
GRADE 3	**130**	22.3
GRADE 4	49	8.4

NON COVID	17	2.9
NORMAL	129	22.2
TB	1	0.2

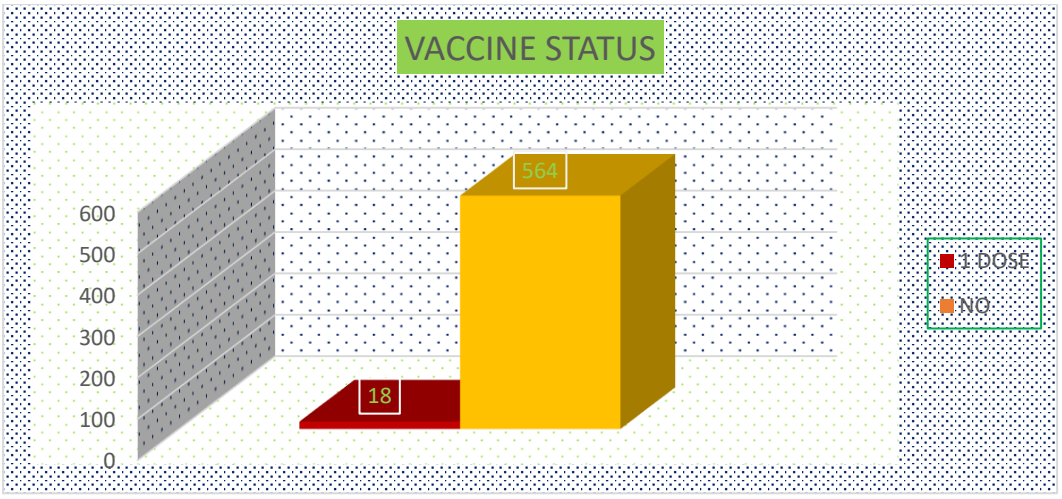
Out of the total study population of 582, 143 (24.6%) had grade 2 lung involvement on CT chest, 111 (19.1%) had grade 1 involvement, 130 had grade 3 (22.3%) lung involvement, 48 (8.2%) had grade 4 lung involvement and 129 had normal findings.

Table No 9 - distribution of steam inhalation among study population

STEAM INHALATION	FREQUENCY	PERCENT
YES	271	46.6
NO	311	53.4

Out of 582 study population 271 (46.6%) gave a history of practicing steam inhalation as a home remedy.

Figure no 10 distributions of vaccination among study population



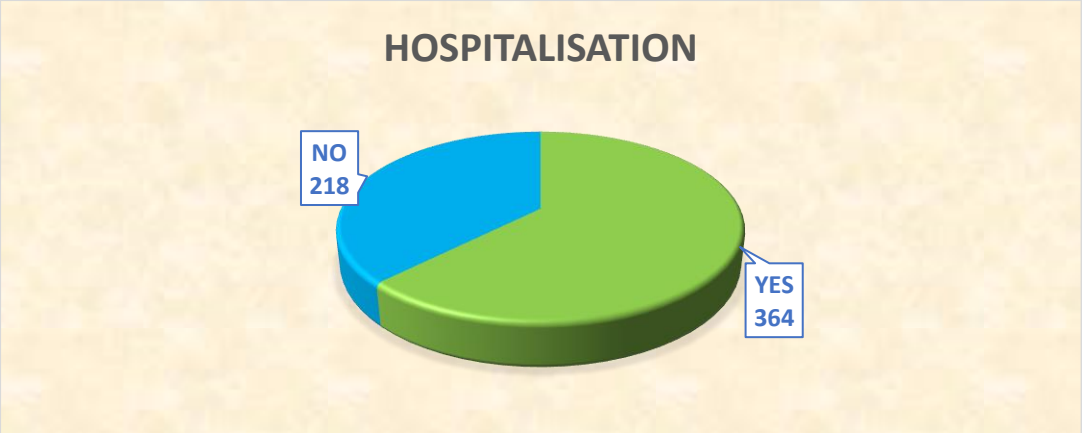
Out of 582 study participants 96.9% were not vaccinated for covid 19. The 3.1% who were vaccinated received only the first dose.

Table no 10 - distribution of Home Isolation among study population

HOME ISOLATION	FREQUENCY	PERCENT
YES	**136**	23.3
NO	446	76.7

136 (23.3%) of the total 582 study population were home isolated.

Figure no 11 distributions of hospitalisation among study population



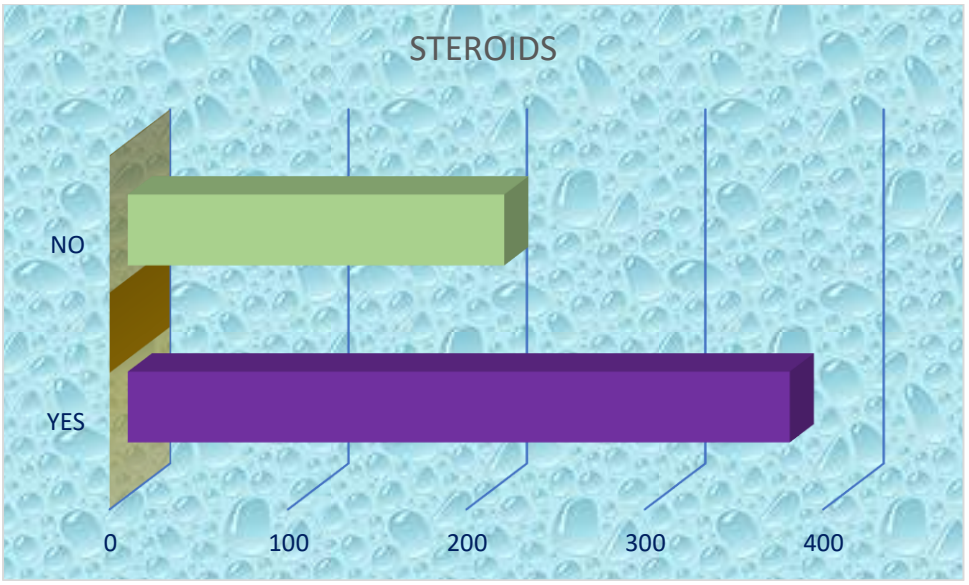
Out of the 582-study population 364 (62.5%) gave a history of hospitalization for Covid-19.

Table no 11- distribution of O2 supply during Hospitalisation among study population

O2 SUPPLY	FREQUENCY	PERCENT
YES	281	48.3
NO	301	51.7

281 (48.3%) participants out of 582 study population needed oxygen during Covid-19 management.

Figure no 12 distributions of steroid intake among study population



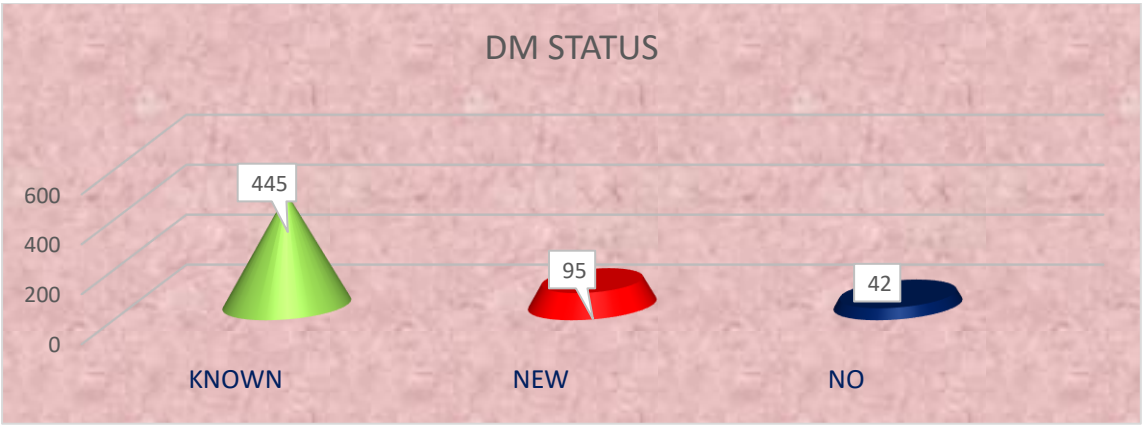
371 (63.7%) cases of the 582-study population were treated with steroids during Covid-19 management.

Table no 12- distribution of zinc supplementation among study population

ZINC SUPLEMENTATION	FREQUENCY	PERCENT
YES	**486**	83.5
NO	95	16.3

Out of the 582 study participants 83.5% had a positive history of prophylactic zinc consumption for covid-19.

Figure no 13- distribution of steroid intake among study population



Out of582 patients studied 445 (76.5%) were known diabetics, 95 (16.4%) were newly diagnosed diabetics and 42 (7%) patients were non-diabetics.

Table no 13- distribution of comorbid condition among study population

COMORBIDITIES	FREQUENCY
TB	4
PLWHA	1
Seizure	1
Hypothyroidism	9
CA Breast CCRT	1
HBSAG	3
Herpes Zoster	1
Coronary Artery Disease	18
Rheumatoid arthritis	1
Chronic Kidney Disease	12
Cerebro Vascular Accident	15
Asthma	3
Chronic lymphoid leukaemia	1
Hodgkins lymphoma	2
ARDS	1
Hypertension	96

146 (25%) cases among the 582 study population had associated comorbidities that include hypertension in 96 cases (16.5%), Cerebro vascular accident in 15 (2.6%), coronary artery disease in 18 (3%), Chronic Kidney Disease in 12 (2.0%), hypothyroidism in 9 (1.5%), Tuberculosis in 4 (0.7%), malignancies in 4 with Chronic Lymphoid Leukemia in 1 (0.2%), Hodgkin's lymphoma in 2 (0.3%) and Post radiotherapy Carcinoma Breast in 1 (0.2%), bronchial asthma in 3 (0.5%), HBsAg in 3 (0.5%), HIV in 1 (0.2%), Herpes Zoster in 1 (0.2%), Rheumatoid arthritis in 1 (0.2%), Acute Respiratory Distress Syndrome in 1 (0.2%) and seizure disorder in 1 (0.2%).

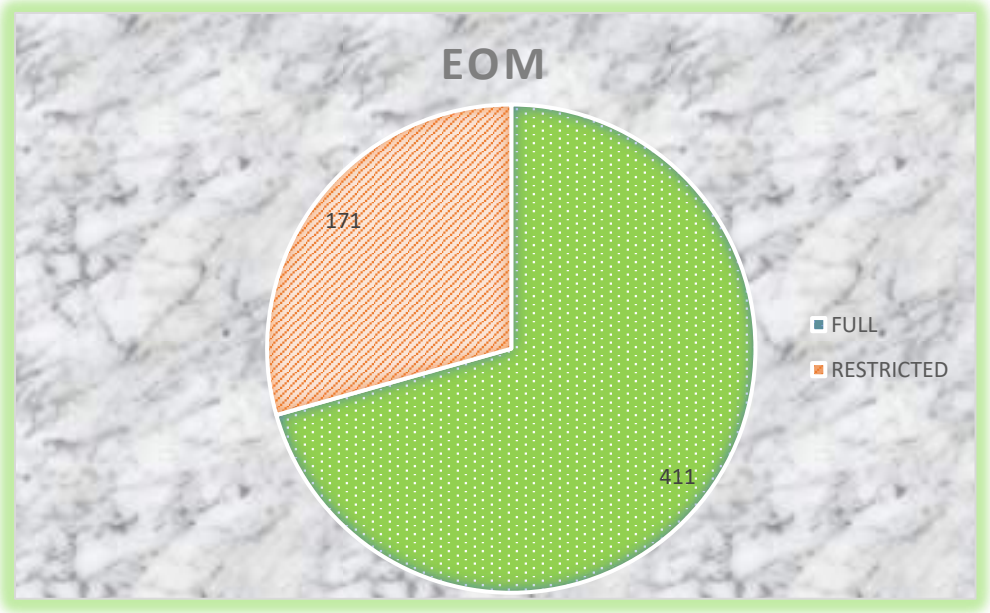
Table no 14- distribution of visual acuity among study population

VISUAL ACUITY	FREQUENCY	PERCENT
Normal	439	75.4
Less Than 1/60	15	10.8
CF	5	0.9
HM	10	1.7
PL	17	2.9
NO PL	94	16.1

Out of 582 cases studied, while 439 (75.4%) had normal vision, 143 (24.5%) had vision disturbances, out of which 94(16.1%) had complete loss of vision, 17 (3%) had diminished

vision (ranging from 6/9 to 1/60), 10 (1.7%) can perceive only hand movements, 17 (2.9%) had perception of light alone, and 5 (0.9%) were able to only count fingers close to face.

Figure no 14- distribution of extraocular movements among study population



In 582 study population, 411 (70.6%) had complete eye movements and 171 (29.3%) had restricted movement of eye balls.

Table no 15- distribution of Ophthalmic examination among study population

OPHTHALMIC EXAMINATION	FREQUENCY
IO anaesthesia	103
Periorbital Edema	87
Chemosis	110
Ptosis	146
Proptosis	53
Lacrimal swelling	2
No	382

Among the 582-study population, 251 (43.1%) had ophthalmological signs that include ptosis in 146 (25%), infra orbital anesthesia in 103 (17.6%) cases, periorbital edema in 87 cases (15%), chemosis in 106 cases (18.9%), proptosis in 53 cases (9.1%) and lacrimal abscess in 2 cases (0.3%). panophthalmitis was detected in 4 cases (0.5%). Majority of the 162 cases had more than one ophthalmological sign on examination.

Table no 16- distribution of Oral examination among study population

ORAL EXAMINATION	FREQUENCY	PERCENT
Ulcer	43	7.3
Bulge	49	8.4
Erosion	58	10
Discolouration	35	6.0
Eschar	16	2.7
Perforation	4	0.7
Oral Thrush	3	0.5
Gum swelling	4	0.7
Gum abscess	1	0.2
Normal	381	65.5

Out of the 582study population, in 201 (34.3%) participants oral involvement was identified in the form of palatal erosion among 58 (10%), palatal ulcer among 43 (7.3%), palatal bulge among 49 (8.4%), palatal discoloration among 35 (6.0%), palatal eschar among 16 (2.7%), palatal perforation among 4 (0.7%), gum swelling among 4 (0.7%), oral thrush among 3 (0.5%) and gingival abscess in 1 (0.2%) participant.

Table no 17 distribution of Diagnostic nasal examination among study population

DNE FINDINGS	FREQUENCY	PERCENTAGE
SECRETIONS	212	36.3
PALE	180	30.9
SLOUGH	114	19.6
ESCHAR	65	11.2
NORMAL	12	2.1

Out of 582 study population, on diagnostic nasal endoscopy, secretions were seen in 212 (36.3%) of participants, pale nasal mucosa was noted in 180 (30.9), slough seen in 114 (19.6%), eschar seen in 65 (11.2%) and normal findings were noted in 12 (2.1%)

Table no 18 distribution of Ophthalmic examination among study population

CT SIDE	FREQUENCY	PERCENTAGE
B/L	262	45
LEFT	157	27.0
NORMAL	5	0.9
RIGHT	158	27.1

Out of 582 study population, 262 (45%) had bilateral sin nasal involvement on CT paranasal sinuses, 157 (27%) showed predominantly left sided involvement and 158 (27.1%) showed predominantly right sided involvement

Table no 19- distribution of Sinuses involved in CT scan among study population

SINUS INVOLVED	FREQUENCY	PERCENT	FREQUENCY	PERCENT
M	46	7.9	59	10.2
E	8	1.4		
F	1	0.2		
S	4	0.7		
ME	87	14.9	116	19.8
MF	7	1.2		
MS	6	1.0		
EF	3	0.5		
ES	13	2.2		
MEF	31	5.3	115	19.8
MES	75	12.9		
MFS	4	0.7		
EFS	5	0.9		
PANSINUSITIS	285	49	285	49
NORMAL	5	0.9	5	0.9
PULMONARY	1	0.2	1	0.2

Among the 582-study population 285 (49%) individuals had pan sinus involvement. Individual sinus involvement was seen in 59 (10.2%), dual sinus involvement was seen in 116 (19.8%). involvement of three sinus cavities was noticed in 115 (19.8%)

Table no 20-distribution of erosion involved in CT scan among study population

EROSION	FREQUENCY	PERCENT
YES	148	25.4
NO	434	75.5

148 (25.4%) of the total study population were reported to have bony erosion on CT paranasal sinuses with orbital cuts.

Table no 21- distribution of Sinuses involved in CT scan among study population

EROSION AT CT SCAN	FREQUENCY
Alveolar Process	88
Maxilla	69
Sphenoid	17
LP	31
Floor of Orbit	7
Ethmoid	5
Frontal	2
Septum	4
Carotid canal	1
No	434

Out of 582 study population, alveolar process was eroded in 88 (15.1%) patients, maxillary walls were eroded in 69 (11.8%) patients, lamina papyracea was eroded in in 31 (5.3%) of patients, sphenoidal erosion (body, greater wing, lesser wing, pterygoid plates) in 17 (3%)

patients, floor of orbit was eroded in 7 (1.2%) of patients, frontal bone in 2 (0.3%), septum in 4 (0.7%), ethmoidal erosion in 5 (0.8%) and carotid canal was eroded in 1 (0.2) patient.

Table no 22-distribution of fat stranding involved in CT scan among study population

FAT STRANDING	FREQUENCY	PERCENT
YES	231	39.6
NO	351	60.4

Out of the582 study population 231 (39.6%) showed evidence of fat stranding.

Table no 23- distribution of Sinuses involved in CT scan among study population

FAT STRANDING	FREQUENCY	PERCENTAGE
EC	143	24.5%
IC	115	19.7%
RETROMAXILLARY	85	14.6%
PREMAXILLARY	56	9.6%
RETROBULBAR	8	1.4%
PERIANTRAL	5	0.9%%
PRESEPTAL	6	1.0%
NO	351	60.3%

Among the 582-study population fat stranding was seen on CT paranasal sinuses with orbital cuts in 39.6% of individuals. Extraconal fat stranding was observed in 143 (24.5%) participants, intraconal fat stranding in 115 (19.7%), retro maxillary fat stranding in 85 (14.6%) participants. Premaxillary in 56 (9.6%) participants, retrobulbar fat stranding in 8 (1.4%) participants, peri antral fat stranding in 5(0.9%) individuals, and Preseptal fat stranding in 6 (1%) individuals. Fat stranding in more than one area is encountered in our study.

Table no 24- distribution of Orbital involvement in CT scan among study population

ORBIT INVOLVEMENT	FREQUENCY	PERCENTAGE
Orbital Invasion	87	14.9
Fat Stranding	159	27.3
Orbit Apex	19	3.3
Orbital Cellulitis	10	1.7
Pre-Septal Abscess	3	0.5
Lacrimal Abscess	2	0.3
No	281	48.4

51.6% of the study population of 582 showed orbital involvement in CT paranasal sinuses with orbital cuts. The multifarious findings noted in orbit on CT were fat stranding (either extraconal intraconal or both) in 159 (27.3%) individuals, orbital invasion (in the form of orbital soft tissue involvement) in 87 (14.9%), orbital apex involvement in 19 (3.3%),

orbital cellulitis in 10 (1.7%), Preseptal abscess in 3 (0.5%) and lacrimal abscess in 2 (0.3%).

Table no 25- distribution of MRI findings in study population

FINDINGS	FREQUENCY	PERCENTAGE
Infarct	18	3
Optic N Involved	16	2.7
CS	44	7.5
ICA	12	2.0
Trigeminal Nerve	6	1
Pre&Retromax Fat	33	5.6
EC&IC Fat	49	8.4
Rectii muscle involvement	6	0.7
Orbital Cellulitis	11	1.9
Orbital Abscess	6	1.0
Orbital Apex	21	3.6
Abscess	23	4
Cerebritis	5	0.9
Granuloma	1	0.2
Meningeal Enhancement	4	0.7
Skull Base Erosion	12	2.0
Superior Orbital Fissure	7	1.2
Others	6	1.0

Among 582 participants, MRI was indicated in 162 individuals only. The results were 18 (11.1%) among the 162 patients with MRI had cerebral infarct. Optic nerve was found to be involved in 16(9.8%) of the MRI44 patients (27.1%) had Cavernous Sinus Thrombosis. Internal carotid Artery was involved in 12 (7.4%) of patients. Trigeminal nerve or Meckel's cave involvement was noted in 6 (3.7%) of study population.33 (20.3%) patients had pre and retro maxillary fat stranding on MRI.49 (30.2 %) showed intra and extraconal fat stranding on MRI. Rectii muscle thickening was noted in 6 (3.7%) of MRI. Orbital cellulitis was present in 11 (6.7%) cases with MRI. In 6 (3.7%) cases MRI showed Orbital abscess. Orbital apex was involved in 21 (12.9%) cases

In 5 (3%) cases MRI showed Cerebritis. Cerebral abscess was noted in 23 (14.2%) cases with MRI Granuloma was seen in 1 (0.6%) of all the MRI.MRI showed Meningeal enhancement in 4 (2.5%) cases. Skull base erosion was noted in 12 (7.4%) of the MRIs in study population Superior orbital fissure was seen to be involved in 7 (4.3%) of the MRIs Other findings like temporal bone osteomyelitis, subperiosteal abscess, b/l periventricular small vessel ischemia and olfactory sulcus involvement were noted in 6 (3.7%) patients

Table no 26- distribution of Stages among study population

STAGES	FREQUENCY	PERCENTAGE	STAGE WISE	TOTAL PERCENTAGE
0	46	7.9	46	7.9
1b	1	.2	2	0.3
1c	1	.2		
2a	23	4.0	235	40.3
2b	22	3.8		
2c	97	16.7		
2d	93	16.0		
3a	88	15.1	218	37.5
3b	46	7.9		
3c	75	12.9		
3d	9	1.5		
4a	10	1.7	81	13.9
4b	16	2.7		
4c	53	9.1		
4d	2	.3		

In our study population of 582 2(0.3%) patients were found to have stage 1 disease involvement, 235 (40.3%) of the patients developed stage 2 disease, 218 (37.5%) developed stage 3 disease and 81 (13.9%) patients developed stage 4 disease.

Table no 27-distribution of KOH mount results among study population

KOH	FREQUENCY	PERCENT
Mucorales	390	67.0
Aspergillus	8	1.3
Rhizopus	2	.4
Mucorales Candida	1	.2
Mucorales Aspergillus	2	.3
Negative	179	30.7

Out of 582 study population 403 (69.3%) turned positive for hyphae in KOH, in which 390 (67.0%) showed Mucorales, 8(1.3%) showed Aspergillus, 3 were mixed with 1 (0.2%) showing Candida and 2 (0.3%) showing Aspergillus in conjunction with Mucorales. No hyphae were detected in 179 (30.7%).

Table no 28- distribution of culture results among study population

CULTURE	FREQUENCY	PERCENT
Mucor	46	7.9
Rhizopus	38	6.5
Aspergillus	10	1.7
Fusarium	1	0.2
Syncephalastrum	1	0.2
No Growth	486	83.5

Out of 582 study population fungal culture was negative in 486 (83.5%) participants. Among the culture positive 96(16.4%) participants, Mucor was detected in 46 (7.9%), Rhizopus in 38 (6.5%), Aspergillus in 10 (1.7%), Fusarium in 1 (0.2%) and Syncephalastrum in 1 (0.2%).

Table no29-distribution of HPE results among study population

HPE MUCOR	FREQUENCY	PERCENT
No	151	25.9
NIM	16	2.8
Angio Invasion	137	23.5
Bone Invasion	9	1.5
Angio Bone Nerve Invasion	1	.2
Angio Bone	256	44.0
Angio Bone Cartilage	2	.3
Mixed Angio Invasion	6	1.0
Mixed Bone Invasion	2	.4
Mixed Angio Bone	2	.4
TOTAL	582	100.0

Out of 582 study population 431 (74%) of them showed Mucor in histopathology while the rest 151 (26%) showed no fungal material in the tissue biopsy. Out of the 431 that are positive HPE for mucor, 1.8% had mixed infection with a combination of Aspergillus or Candida or both. Out of the 431 patients showing fungal hyphae on HPE 415 showed signs of invasion while the rest, 16 patients had noninvasive Mucormycosis. Different types of

invasions that were observed include angio invasion in 137 (23.5%), both angio and bony invasion in 256 (44%), bone invasion alone in 9 (1.5.%), cartilage invasion in 2 (0.3%) and nerve invasion in 1 (0.2%)

Table no 30-Association between histopathology and KOH mount

	HPE POSITIVE	HPE NEGATIVE
KOH POSITIVE	53.6%	15.4%
KOH NEGATIVE	20.6%	10.4%

In our study of 582 population both KOH and HPE were positive among 313(53.6%) individuals, both were negative in 61(10.5%), HPE was positive among 118(20.6%) of individuals with a negative KOH and KOH was positive in 90 (15.4%) of individuals who were negative for HPE.

ASSOCIATION WITH MUCORMYCOSIS

Table no 31 – association between Facial pain and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
FACIAL PAIN	YES	243 95.3%	12 4.7%	6.376	2.350	1.191	4.637	**0.012**
	NO	293 89.6%	34 10.4%					

In this table we calculated the association between facial pain and Mucormycosis and the p value is -0.01.so there is significant association existing between facial pain and Mucormycosis

Table no 32– association between Facial swelling and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
FACIAL SWELLING	YES	229 94.6%	13 5.4%	3.648	1.894	0.975	3.679	0.05
	NO	307 90.3%	33 9.7%					

In this table we calculated the association between facial swelling and Mucormycosis and the p value is - 0.05.so there is significant association existing between facial swelling and Mucormycosis

Table no 33– association between nasal block and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
NASAL BLOCK	YES	454 93%	34 7%	3.641	1.954	0.971	3.931	0.05
	NO	82 87.2%	12 12.8%					

The above table shows that 93. % of the study population with nasal obstruction were associated with Mucormycosis. Significant association was observed between nasal obstruction and Mucormycosis (0.05).

Table no 34– association between mucoid discharge and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
MUCOID NATURE	YES	361 93.8%	24 6.2%	4.358	1.891	1.032	3.466	0.03
	NO	175 88.8%	22 11.2%					

The above table shows that 93.8%% of the study population with mucoid nasal discharge were associated with Mucormycosis. Significant association was observed between mucoid nasal discharge and Mucormycosis (0.03).

Table no 35– association between covid status and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
COVID STATUS	POST COVID	465 93%	35 7%	3.982	2.058	1.000	4.237	0.04
	NON COVID	71 86.6%	11 13.4%					

The above table shows significant association between Covid-19 infection and development of Mucormycosis (P 0.04).

Table no 36 – association between Hospital admission and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
HOSPITAL ADMISSION	YES	315 94.9%	17 5.1%	8.226	2.431	1.304	4.533	0.03
	NO	221 88.4%	29 11.6%					

The above table shows significant association between hospitalization during Covid-19 and development of Mucormycosis (P 0.03).

Table no 37– association table between other variables and mucormycosis occurrence

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
SEX	MALE	377 91.5%	35 8.5%	0.678	0.745	0.369	1.504	0.410
	FEMALE	11 6.5%	159 93.5%					
FACIAL NUMBNESS	YES	66 97.1%	2 2.9%	2.605	3.089	0.732	13.043	0.17
	NO	470 91.6%	44 8.6%					
EYE PAIN	YES	171 94.5%	10 5.5%	2.042	1.687	0.818	3.478	0.15
	NO	365 91%	36 9%					
EYE SWELLING	YES	140 91.5%	13 8.5%	0.100	0.897	0.459	1.754	0.75
	NO	396 92.3%	33 7.7%					
VISUAL DISTURBANCES	YES	108 89.3%	13 10.7%	1.693	0.641	0.326	1.249	0.19
	NO	428 92.8%	33 7.2%					
HEADACHE	YES	245 92.8%	19 7.2%	0.332	1.996	0.649	2.204	0.56
	NO	291 91.5%	27 8.5%					

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
TOOTHACHE	YES	76 90.5%	8 9.5%	0.354	0.785	0.353	1.747	**0.55**
	NO	460 92.4%	38 7.6%					
BLOOD-STAINED NASAL DISCHARGE	YES	44 93.6%	3 6.4%	0.162	1.282	0.382	4.300	**0.62**
	NO	492 92%	43 8%					
TOOTH LOOSENING	YES	18 90%	2 10%	0.125	1.308	0.294	5.821	**0.72**
	NO	518 92.2%	44 7.8%					
STEAM INHALATION	YES	255 94.1%	14 5.9%	2.786	1.702	0.906	3.195	**0.09**
	NO	281 90.4%	30 9.6%					
HOME ISOLATION	YES	151 88.8%	19 11.2%	3.534	0.557	0.301	1.032	**0.07**
	NO	385 93.4%	27 6.6%					

VARIABLE	RISK	MUCOR	NO MUCOR	χ^2 VALUE	OR	MINIMUM	MAXIMUM	P-VALUE
TOOTHACHE	YES	76 90.5%	8 9.5%	0.354	0.785	0.353	1.747	**0.55**
	NO	460 92.4%	38 7.6%					
ZINC SUPPLY	YES	452 92.8%	35 7.2%	2.107	1.691	0.826	3.462	**0.16**
	NO	84 88.4%	11 11.6%					

In this table association between Sex, facial numbness, eye pain, eye swelling, visual disturbances, headache, toothache, bloody nasal discharge, loosening of tooth, steam inhalation, zinc consumption and mucor was calculated and there was no association detected between the variables and mucor occurrence.

DISCUSSION

Demography

Mean age of CAM affliction in our study was found to be **52.69 ± 10.98** year with a male predominance (70.8%), which was consistent with studies by Sen et al., 2021 (51.9% with 71% males) [13] and Mishra et al.,2021; (58.28 ± 8.5 with 53.1% males) [76]. we found that the prevalence of CAM to increase gradually with age, peaking in fifth decade and decreasing thereafter.

Clinical features

DURATION:

The average **duration** of onset of Mucormycosis from day of positivity was found to be **13±22.05** days in our study.

Muthu et al.,2021; observed that on an average CAM was reported after approximately 19.5 days from the date of covid-19 positivity [78].

Mishra et al.,2021; remarked 17.28 (±11.76 day) to be the mean duration of CAM diagnosis from the date of Covid positivity. [76]

Sen et al., 2021; determined the mean interval to be 14.5 ± 10 days [13].

The maximum duration of onset of Mucormycosis from the day of Covid positivity was 6months which highlights that post covid patients need to be followed up for a **maximum of 6 months** to look for signs of Mucormycosis.

RISK FACTORS OF CAM

COVID-19:

85.9% of our study population were afflicted by **Covid-19** during the second wave, out of which 93% developed Mucormycosis.

A strong association was found in our study between **Covid-19 (P 0.04)** and Mucormycosis suggesting Covid-19 to be an important underlying factor for developing Mucormycosis.

56.2% of the patients with Mucormycosis had a history of Covid -19 in a study conducted by Mishra et al.,2021; and he found it to be significantly associated with CAM [(76)].

Among Our study population 43.7% of covid infected individuals showed less than 50% lung involvement and 30.5% of Covid infected individuals had more than 50% lung involvement. 22.1% of individuals in our study with a history Covid-19 showed no lung involvement during the infection.

The findings were similar to a study conducted by Abdel-Tawab.,2021; on CT chest severity in Covid-19 who identified 47.9% of study population to have less than 50% lung involvement and 23.1% more than 50% lung involvement [(94)].

HOME ISOLATION & HOSPITALISATION DURING COVID:

In our study, out of 500 participants who had a history of Covid-19 infection, 72.8% were hospitalized and 27.2% home isolated, similar to that of Sen et al.,2021; who reported that

28% of the study group were home isolated and 72% required hospitalization for Covid-19 treatment [13].

In our study 94.9% of participants with a history of hospitalization during Covid-19 infection developed CAM and a significant association was found between **hospitalization (P 0.03)** and Mucormycosis.

Patel et al., 2021; found the incidence of CAM to be 65.2% and prevalence to be 0.27% in hospitalized covid patients [68].

STEROID ADMINISTRATION:

74.2% of study population received steroids for Covid-19 management in our study while Sen et al., 2021; described 87% [13] and Mishra et al.,2021 detected steroid administration in 93.7% of CAM patients [76]

In our study 93.8% who received steroids during Covid developed Mucormycosis and a significant association **(p=0.04)** was found between the two.

One of the mainstay treatments in the management of covid-19 include corticosteroids like dexamethasone and methylprednisolone that were known to interfere with the immune systems eradication of fungi [68]. and their tendency to predispose to elevated blood sugar levels together may have been the possible reason behind it.

DIABETES:

92.9% of our study population were diabetics, the major risk factor seen in our study underlying Mucormycosis. This is consistent with the studies of pre covid era, like roden et al.,2005;

It also accords with in post covid era studies like Muthu et al.,2021; who remarked **Diabetes mellitus** to be the most frequent underlying risk factor for CAM in India (66.1%) when compared to the rest of the world with (54.8%) [54]

Sen et al., 2021;(78%) [13] and Mishra et al.,2021; (87.5%) [76]. concluded that diabetes is a major risk factor for ROCM.

The most common type of CAM reported in our study is Rhino-Orbito-Cerebral form and was found to be associated more commonly with Diabetes.

This is in accordance with a study by Jeong et al.,2019, who stated rhino cerebral Mucormycosis to be commonly associated with diabetes [15].

Though traditionally Mucormycosis was associated with diabetic keto acidosis, we encountered only 2.1% of patients with Diabetic keto acidosis in our study

Out of 540 of our study population who were diabetics, 95 (17.6%) were new onset diabetes mellitus developed post covid infection.

Sathish & Chandrika Anton et al.,2021; pointed out that 20.6% of post covid patients had new onset diabetes mellitus [77].

COVID-19 VACCINATION STATUS:

In our study 96.9% of the individuals were not vaccinated and the rest (3.1%) were partially vaccinated with a single dose

In a study conducted by Mishra et al.,2021; none of the study population were fully **vaccinated** against Covid while two of them received first dose **[76].** Hicks et al.,2021; reported 89% [77] and Shankar et al.,2021 remarked 86% [76] of the Mucormycosis patients to be unvaccinated.

These findings hint at the efficacy of vaccine in protecting against Covid-19 and CAM.

OXYGEN THERAPY:

In our study population 56.2% of the covid affected participants required oxygen therapy. In a study by Sen et al., 2021; 57% were on **oxygen** support for covid-19 during hospitalization. [13]

Our study found no significant association between oxygen therapy and Covid-19.

Muthu et al., 2021; felt that industrial oxygen has no role in the surge of CAM [54]. Kumar et al.,2021; suspected that industrial oxygen could be a contributing factor for the epidemic [35].

Aranjani et al.,2021; Gupta et al.,2021; debated that Mucorales lacks the ability to produce spores in water of humidifiers agitated by the passage of oxygen [80,81].

STEAM INHALATION:

46.6% of our study population had a history of steam inhalation, but our study showed no significant association between steam inhalation (P 0.09) and CAM.

ZINC SUPPLEMENTATION:

Though 83.5% of our study population consumed zinc as a prophylaxis against Covid-19, our study found no statistical significance between zinc consumption & Mucormycosis.

Our results accord with that of Muthu et al.,2021; who found no conclusive evidence suggesting that zinc supplementation contributed to Mucor mycosis epidemic [78].

OTHER ASSOCIATED COMORBIDITIES:

In our study we found hypertension (16.5%) to be the second most commonly associated comorbidity after diabetes.

Apart from diabetes, hypertension was the most commonly associated comorbidity with CAM in studies by Sen et al., 2021 (80%) [13]; and Mishra et al.,2021;(50%) **[76]**.

We observed haematological malignancies (chronic lymphoid leukaemia and Hodgkin's lymphoma) in only 3 (0.5%) of the study population with all three of them being post covid and diabetics.

These results are as seen in a study by Hussain et al.,2021; that showed only 0.89% of CAM patients to be associated with underlying haematological malignancy [6].

Our study determined that 2.6% and 3% of the study population to be associated with CVA and CAD respectively suggesting the reason to be the hypercoagulable state of Covid-19.

Sen et al.,2021; detected cardiovascular disease in 1.9% and cerebrovascular disease in 0.9% of Post covid patients with Mucormycosis.

In our study population **25 (4.3%) of cases had no associated comorbidities except for Covid-19** and 6 (1%) of them had no history of oxygen therapy or steroid administration during their Covid treatment.

Sen et al.,2021; identified that 18% of the CAM patients had no underlying comorbidities apart from Covid -19 and 5% (23) of them received neither O2 nor corticosteroids.

This stresses the need to have a high index of suspicion for Mucormycosis and to carefully investigate for the same in any patient presenting with features of sinusitis in the background of Covid-19 with no other risk factors and highlights the need for further studies to thoroughly understand the interactions of Covid-19 and its role as the potential sole risk factor for CAM.

It is of note that 6(1%) of our total study population with CAM had no history of Covid-19 infection and were neither diabetics nor had any other comorbidities. The possibility of a subclinical Covid-19 infection couldn't be ruled out in these patients hinting at the hidden clinical burden of hidden Covid-19 infections in the community.

CLINICAL PRESENTATIONS:

SYMPTOMS:

In our study population presented with a myriad of **symptoms** with predominantly nasal obstruction (83.8%), nasal discharge (66.2%), facial pain (43.8%) and facial swelling (41.6%) closely followed by headache (45.4%). the other symptoms noted were ocular symptoms like vision disturbance (20.8%), eye swelling (26.3%) and eye pain (31.1%). Skin involvement was noted in our study in the form of facial discoloration progressing to facial skin necrosis and eschar formation over the face. In our study it was found that out of all the symptom, nasal obstruction (0.05), nasal discharge (0.03), facial swelling (p 0.05), facial pain (p 0.012) were the clinical features to be significantly associated with Mucormycosis.

Though initially we encountered patients with late-stage symptoms predominantly, with the public education strategy and initiation of Mucormycosis Screening Clinic by the Government of Tamilnadu, patients started to seek medical treatment with initial warning symptoms itself.

Various symptoms of CAM observed by Sen et al., 2021; include orbital or facial pain in 23%, facial or orbital Edema in 21%, vision loss in 19% and nasal stuffiness in 9% [13]

Mishra et al.,2021; reported headache in 93.8% of his study population, nasal blockage & discharge in 62.5% and ocular symptoms in 59.4% as presenting features of CAM [76].

In our study we had 21 (3.6%) patients with facial palsy which is similar in findings to the studies by Sachdev et al.,2013; and Mehta et al.,2021; (2%)[96].

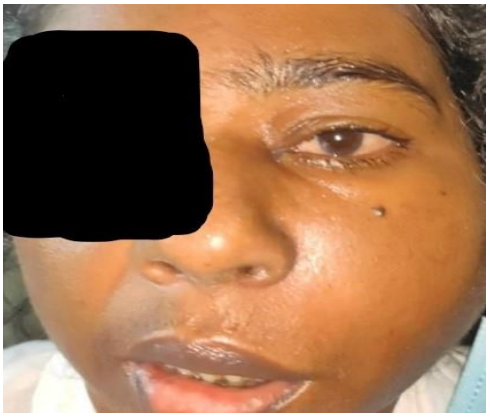
Apart from CVA, over the background of post covid status and diabetes, one should be wary of the possibility of Mucormycosis in patients presenting with facial palsy.

Figure no:15 Nasal discharge symptoms in patients admitted in RGGGH

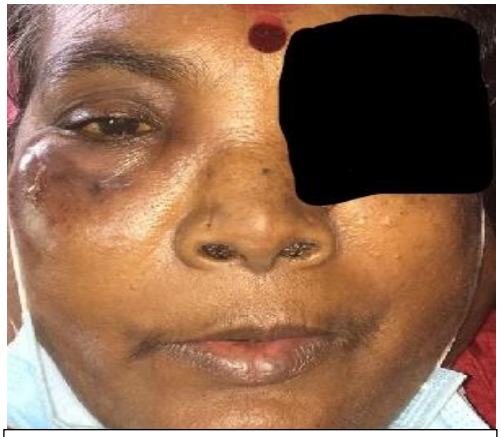
Blackish nasal discharge from right nose

Mucopurulent discharge from right nostril

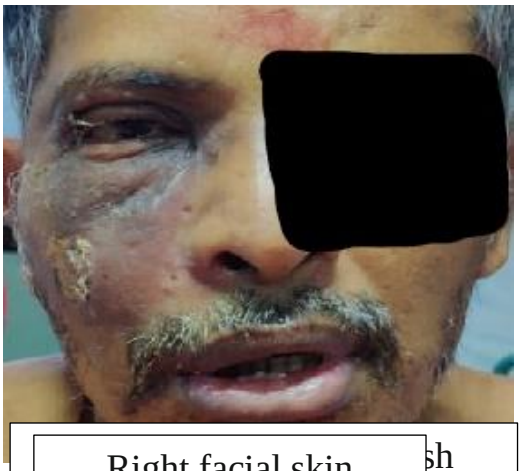
Figure No 16-Picture of facial swelling Taken from RGGGH, mucor ward



LEFT FACIAL SWELLING

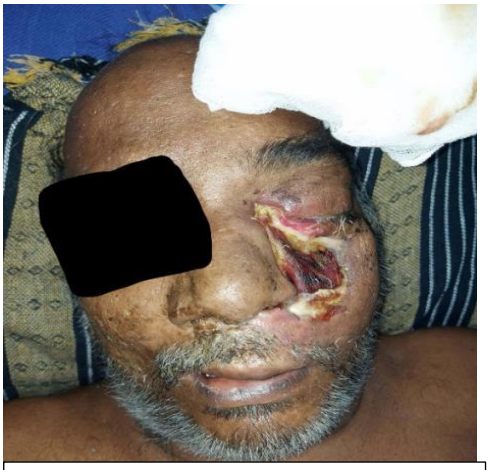


Right facial skin early discoloration



Right facial skin blackish discoloration

Early Necrosis of left facial skin



Necrosis of left facial skin

Eschar over right facial skin


Right Facial Nerve Palsy

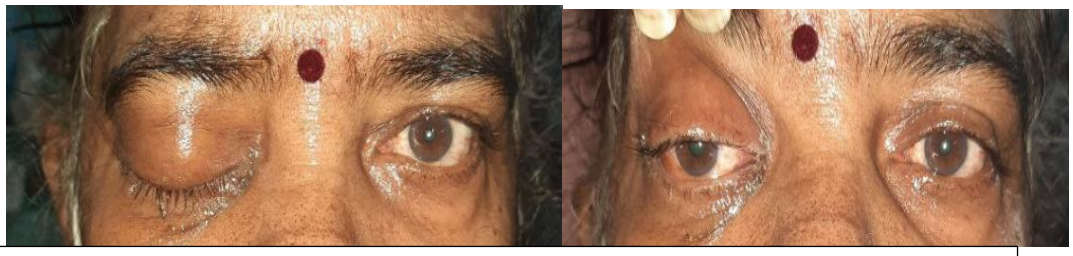
OPHTHALMOLOGY SIGNS:

Restriction of ocular movements (29.3%) and Ptosis (25%) were the most common ophthalmological signs noted in our study population followed by chemosis (18.9%) & Infra-orbital anesthesia (17.6%). Abduction restriction was noted to be the earliest sign of orbital involvement in our study appearing even before ptosis, periorbital edema and diminished vision.

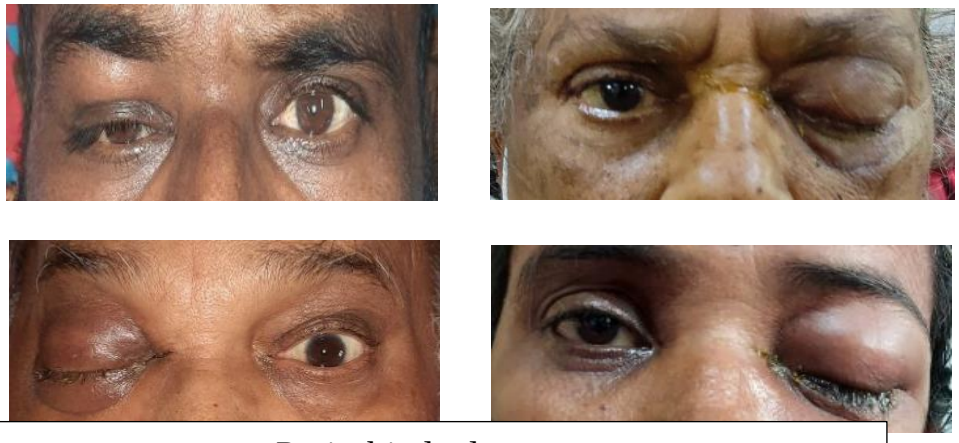
143 (24.5%) of our study participants presented with visual disturbances and among them 94 (16.1%) had complete loss of vision. Other signs noted in our study include periorbital edema (15%), chemosis (18.9%) and proptosis (9.1%) . Panophthalmitis, one of the severe forms of orbital Mucormycosis, was noted in 0.5% of our study population. Rare presentation of Lacrimal abscess was observed in 0.5% of cases.

Signs of CAM noticed by Sen et al., 2021; were periorbital edema (33%), loss of vision in (21%), ptosis in (12%) and proptosis in (11%) [(13)].

Figure No:17 Pictures showed periorbital edema taken from RGGGH



Early orbital involvement - Right Eye Ptosis with No Congestion or Chemosis



Periorbital edema

Right eye Chemosis & Congestion

Figure No: 18 pictures showed Ptosis in mucor Patients taken from RGGGH

Right eye Ptosis & severe Chemosis

B/L eye ptosis, Proptosis & severe Chemosis

Left eye Ptosis, Proptosis, Periorbital edema with Discharge

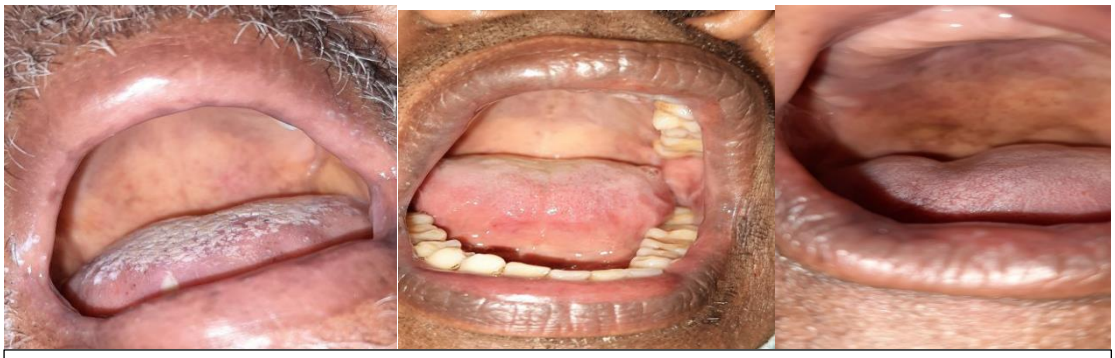
ORAL SIGNS:

201 (34.3%) of our study population had oral manifestations that include palatal erosion (10%), palatal bulge (8.4%), ulcer (7.3%), palatal discoloration (6%), palatal erosion (2.7%) and palatal perforation (0.7%).

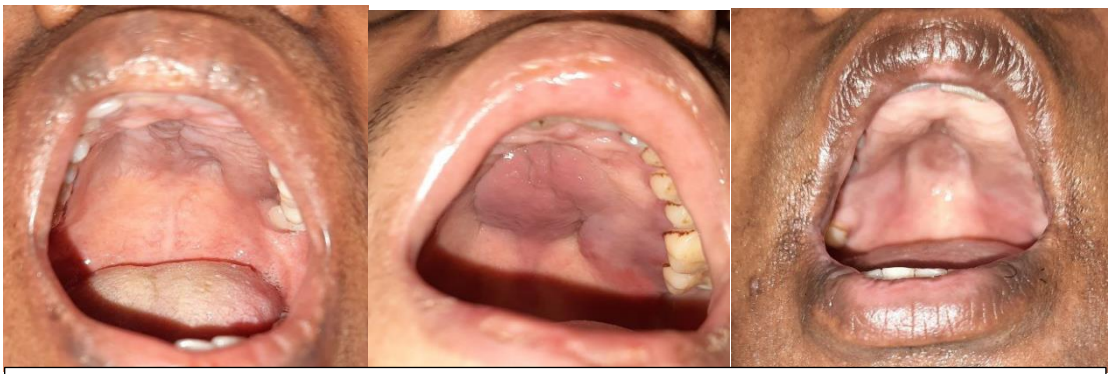
Jones et al.,1993; observed palatal ulcer to be the most frequent oral sign in Mucormycosis [(92)]. Ribes et al.,2000; reported palatal necrosis to be caused by extension of Mucor from sinuses [(31)]. Blackish palatal eschar was noted in Mucormycosis patients by Jain et al.,2014[(85)]. In post covid era Mitra et al., 2021; observed palatal involvement in 25% of CAM patients [(91)]

Sharma et al.,2021; reported blackish eschar over hard palate as oral manifestation in 39% of patients [90].

Figure No: 19 pictures showing palatal changes in mucor patient taken from RGGGH.



Palatal
Palor and Discoloration



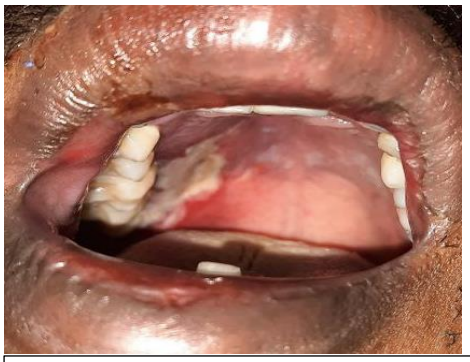
Palatal Bulge

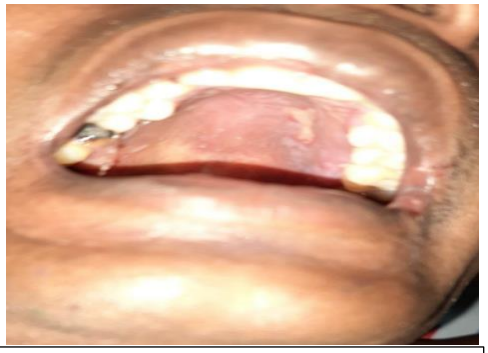
0.4% patients in our study presented with palatal bulge alone as the presenting sign indicating the need for high index of suspicion in diagnosing Mucormycosis.

A literature review by Nicolatou-Galitis et al.,2015; found palatal signs to be the sole presenting complaint in 8 (0.6%) patients in his study leading to the diagnosis of Mucormycosis [86]As the presenting complaint and being easily accessible ulceration of the hard palate aids in early diagnosis of Mucormycosis [86] [93].

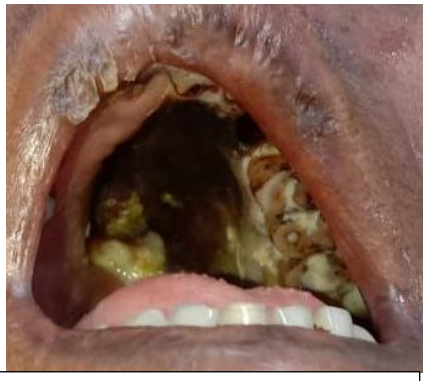
Gingival swelling was encountered among 4 participants and gingival abscess was seen in one individual in our study.

Figure No: 20 pictures showing palatal ulcer taken from RGGGH

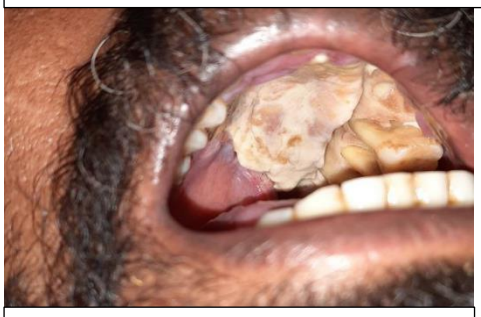




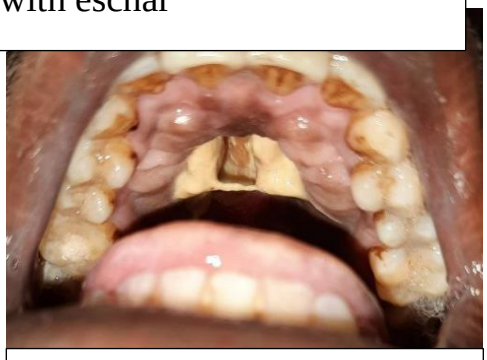
Palatal Ulcer



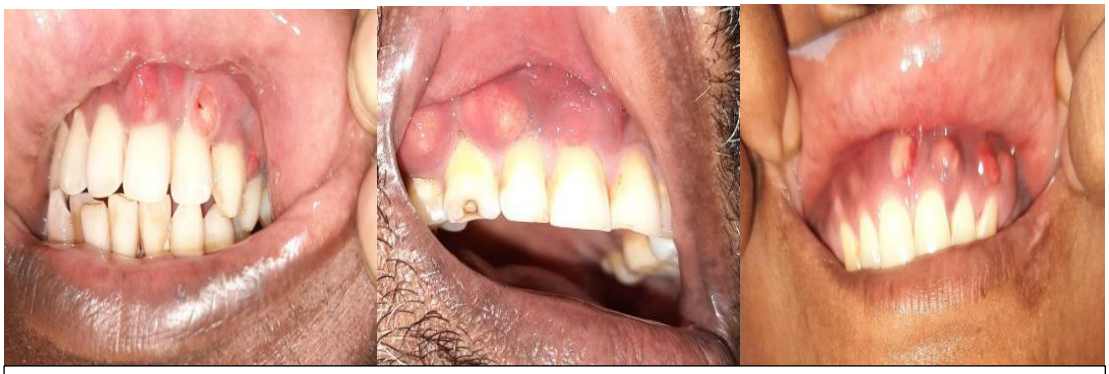
Palatal Necrosis with eschar



Palatal Erosion



Palatal Perforation



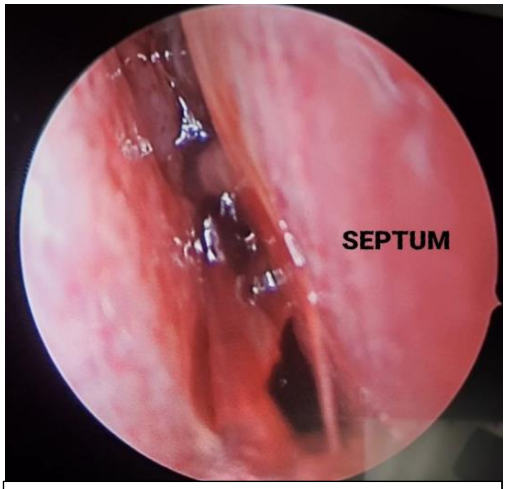
Gingival Swellings

Diagnostic investigations:

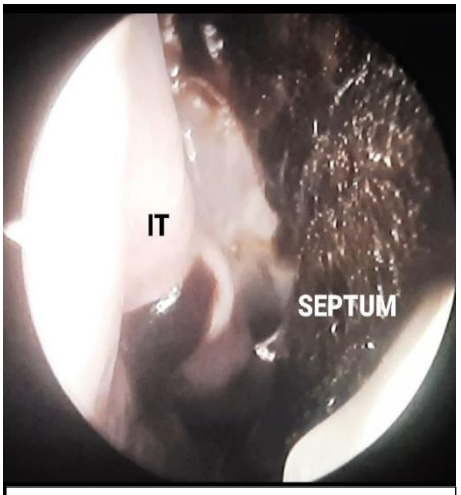
DIAGNOSTIC NASAL ENDOSCOPY:

. Our study identified mucosal pallor (30.9%) in most of the study participants with dirtyslough seen in 19.6% and eschar seen over turbinate and septum in a 11.2% of participants. Normal DNE findings were noted in 2.1% of our study population.

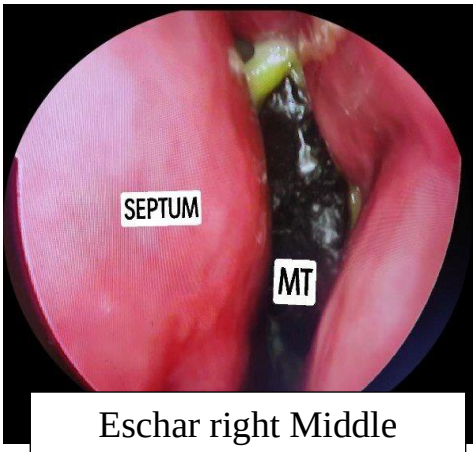
Singh et al.,2019; noticed mucosal discoloration in early stages and blackish eschar involving middle turbinate, septum and inferior turbinate in his study [107].



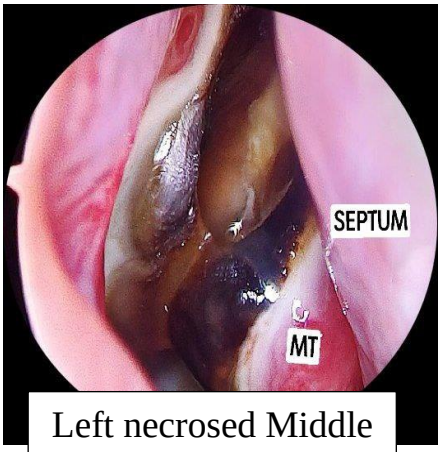
Right blood-stained nasal secretions



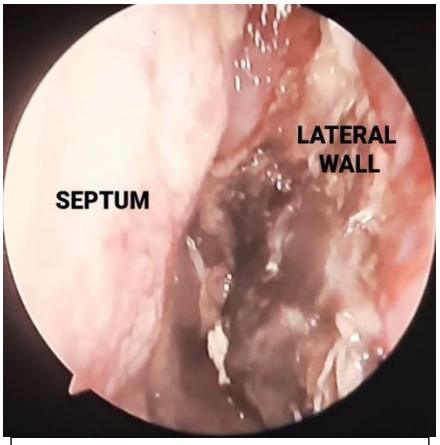
Eschar - Nasal Septum



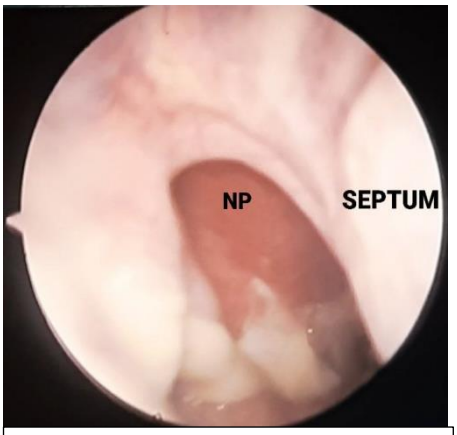
Eschar right Middle Turbinate & Ethmoid



Left necrosed Middle Turbinate



Necrosed lateral wall of left Nasal Cavity

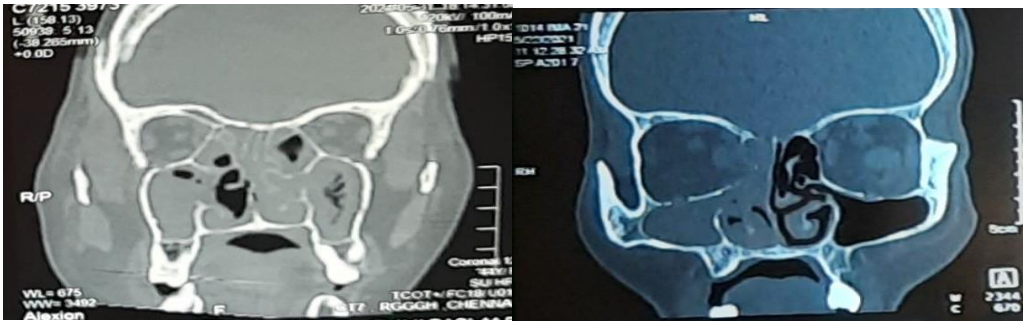


Right choana with purulent secretions - secondary bacterial infection

RADIOLOGY: CT

Laterality -

our study noted bilateral sinus involvement radiologically in 45% of individuals and 54% showing unilateral sinus involvement among which side predominance was seen equally on both sides (27%.) which is consistent with findings of Sen et al., 2021;(unilateral in 59% and bilateral in 40% [13]).



CT.**A.** CT. **B.**

Fig.A. 21 Shows Coronal CT with bilateral maxillary and posterior ethmoid sinus opacification and **B.** shows opacification of unilateral right Maxillary & Ethmoid sinus opacification

Sinus predominance-

In our study Maxillary sinus was the most frequently involved sinus followed by ethmoid and sphenoid with frontal being the least involved.

Studies by Sen et al., 2021; Desai et al.,2021; Mishra et al.,2021; & joshi et al.,2021 reported maxilla to be the frequently affected sinus followed by ethmoid [13][97]**[76]** [103]. Sharma et al.,2021 noted ethmoid to be the most common sinus to be involved followed by maxilla [90].

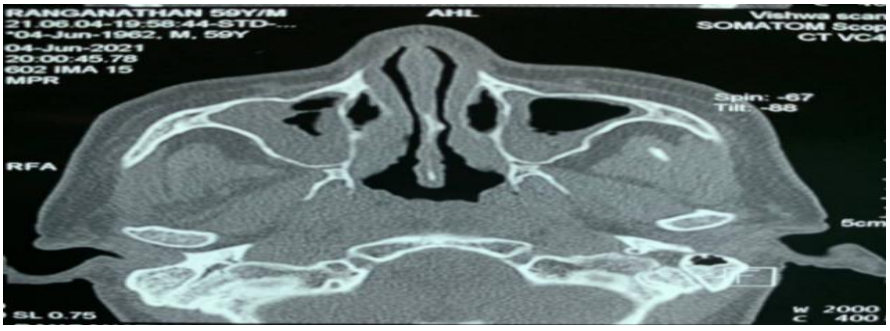


Fig. 22 Shows Axial CT with soft tissue opacification of the bilateral maxillary sinuses and normal pre and retro antral fat

Orbital findings in CT:

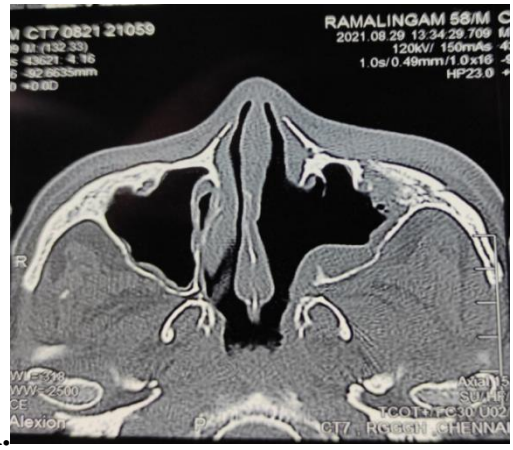
In our study Orbital involvement was discerned on CT in 51.6% participants of our study population with the manifestations ranging from early signs like orbital fat stranding to advanced stage signs like orbital cellulitis. 27.3% of total cases had fat stranding in intra & extraconal regions with involvement of orbital soft tissues in Preseptal/retrobulbar seen in 14.9% individuals.

Mishra et al.,2021; observed orbital involvement in 59.4% CAM cases radiologically [(76)].

Bony erosion in CT:

Bony erosion, a sign of advanced Mucormycosis, was encountered in 25.4% of our study population with majority seen in maxilla (11.8%) and alveolar process (15.1%). Other bony erosions encountered were sphenoid, lamina papyracea, frontal and ethmoids.

Joshi et al.,2021; reported bony erosions in 30% of his study population involving sinus walls, nasal septum, ethmoids, orbital walls, cribriform plate, wings of sphenoid, Sella, skull base and hard palate [(104)].

A. 

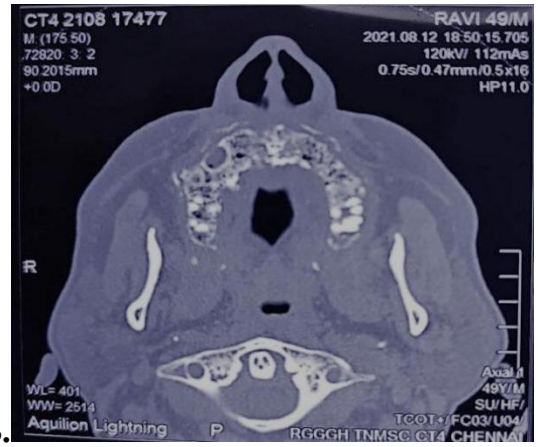
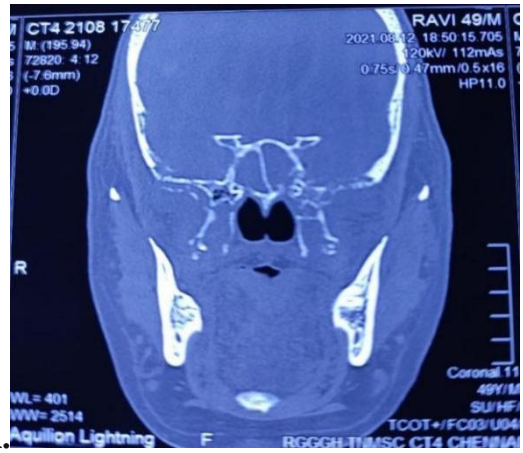
B. 

Fig A. 23 shows Plain CT axial cut with porosity and erosion of left anterolateral wall of maxilla and **B.** Axial CT shows erosion of bilateral alveolar process

A. 

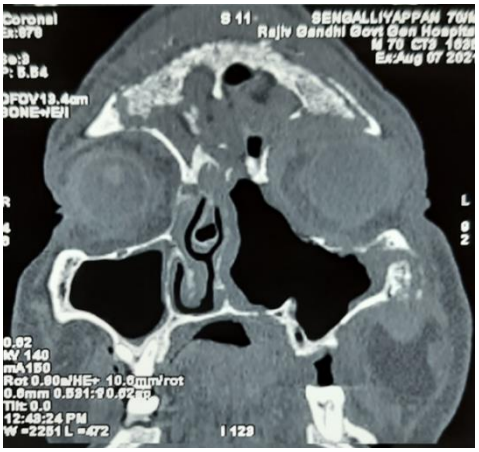
B. 

Fig A. shows Coronal CT with erosion of bilateral greater wing of sphenoid, pterygoid wedge & pterygoid plates **B.** Plain CT coronal cuts showing erosion and sequestrum with subperiosteal abscess frontal bone

Fat stranding in CT:

Though MRI was known to be more sensitive and superior to CT in detecting soft tissue changes in extra sinus tissues like fat stranding [51][53] we were able to identify fat stranding in 39.6% of our study population involving intraconal space (19.7%) extraconal space (24.5%), premaxillary space (9.6%)/ retro maxillary space (14.6%), peri antral space (0.9%) and retrobulbar space (1.4%), a more specific finding to invasive Mucormycosis.

Earlier studies in the Precovid era, the likes of Green et al.,1967; Addlestone et al.,1975; & Gamba et al.,1986; mainly highlights the bony erosive nature of Mucormycosis [101] [102] [103]. As bony destruction is seen mostly in advanced stages of Mucormycosis, later studies of Silverman & Mancuso.,1998; and DelGaudio et al., 2003; focused on CT changes that appear much earlier in the course of the disease stressing on the invasive nature of Mucormycosis like extra sinus soft tissue involvement in the form of fat stranding. [99] [100].

Thus, the diagnostic value of CT in detecting early signs was significant.

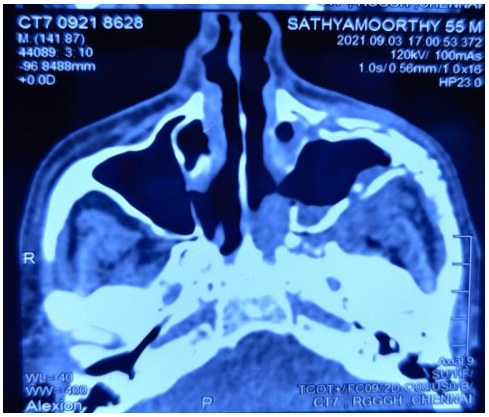
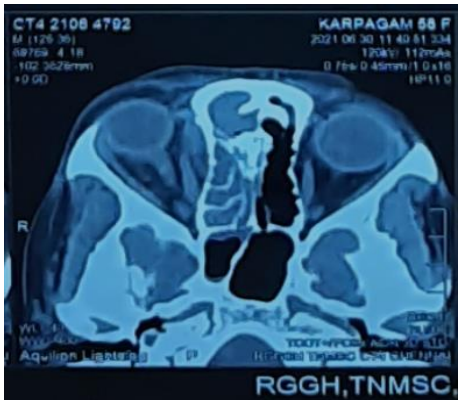


Fig 24 shows Axial CT with left Pre & Retro maxillary fat stranding with erosion of anterolateral & posterolateral walls of left maxilla

A. 

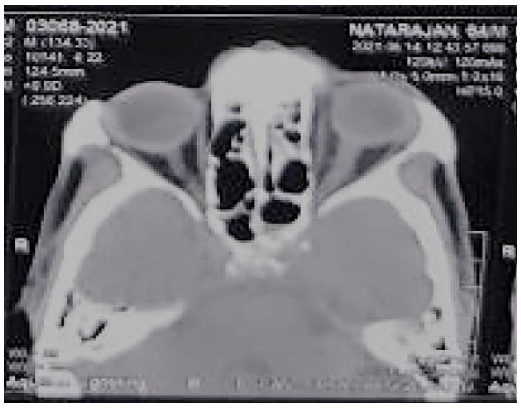
B. 

Fig 25 A. shows Axial CT with extraconal fat stranding right side with unilateral ethmoid sinus opacification and **B** Axial CT showing intraconal fat stranding on left side.

In our study we noticed evidence of soft tissue involvement on CT without involvement of bony boundaries in 214(36.7%) of the participants.

Aribandi et al.,2007; described cases where involvement of bone could be insignificant and extra sinus involvement might be appreciated even with intact bony walls of the sinuses and orbit owing to the Angio invasive nature of the Mucoralean fungi [98].Desai et

al.,2021;noted that extra sinus soft tissue hypo intensity could be evident even with no bony erosion [97].

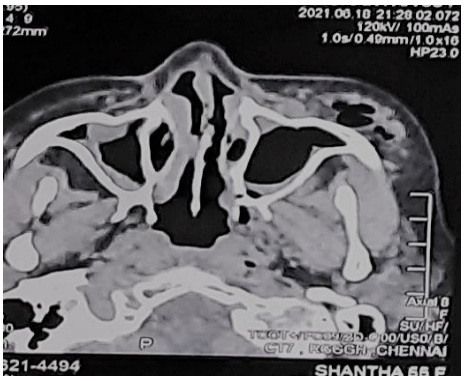


Fig 26 shows Axial CT switch left pre & retro maxillary fat stranding with air pockets in left premaxillary region with intact bony walls

In our study we noticed 96(16.4%) cases with no evidence of soft tissue invasion or bony invasion on CT, out of which 12 individuals were proven to have noninvasive Mucormycosis on HPE and the rest, 84 individuals showed evidence of angio and bone invasion on HPE.

RADIOLOGY: MRI

In our study MRI was taken in 162 participants and the findings showed hypo intensity of involved tissues including classical signs like black turbinate sign. Desai et al.,2021; observed that fungal involvement in CAM to appear as hypo intensity on MRI which can be classically seen as Black Turbinate sign[97].

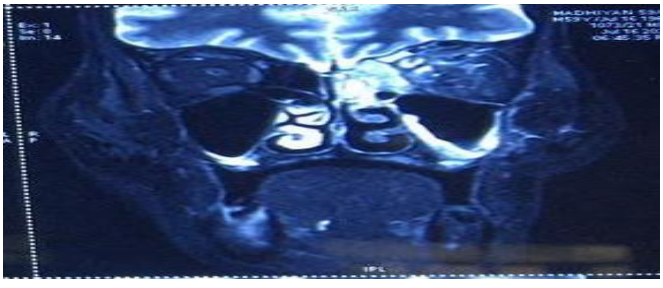


Fig. 27 T1 contrast enhanced MRI coronal and axial cuts showing focal lack of enhancement of the left nasal turbinate mucosa -'Black turbinate sign' - an Early Sign.

Orbital invasion MRI -

In our study Orbital invasion on MRI was evident in the form of fat stranding in 8.4% patients, orbital apex involvement was noted in 3.6% patients, orbital cellulitis was seen in 1.9% patients, orbital abscess in 1.0% cases, Rectii muscle involvement was seen in 1.0% of patients and superior orbital fissure soft tissue involvement in 1.2%. of total study population. Diffuse orbital involvement with severe proptosis and increases intraorbital pressure may tent the globe posteriorly called as "**guitar pick sign"** [(108)] was encountered in our study.

Desai et al.,2021; reported that Orbital invasion on MRI can be observed in the form of muscle thickening, extra & intraconal fat stranding, optic nerve involvement and orbital apex soft tissue involvement [(97)]

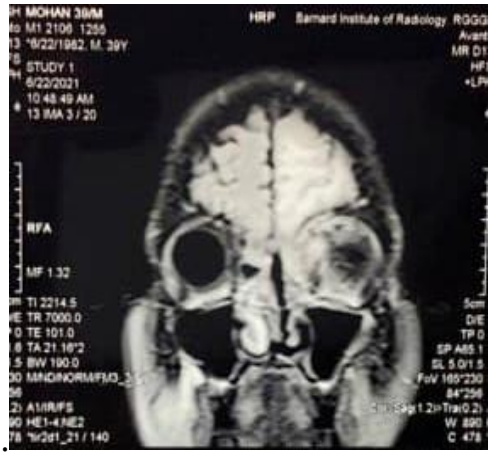


Fig. 28 T1 weighted MRI shows soft tissue thickening in intra & extraconal space of left orbit

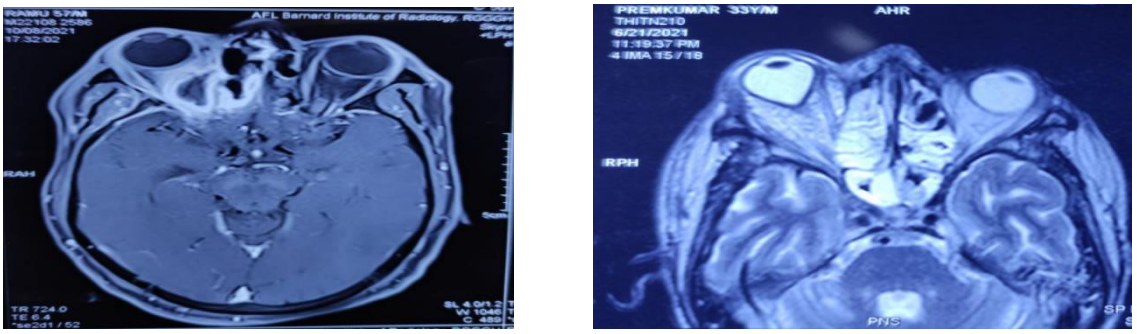
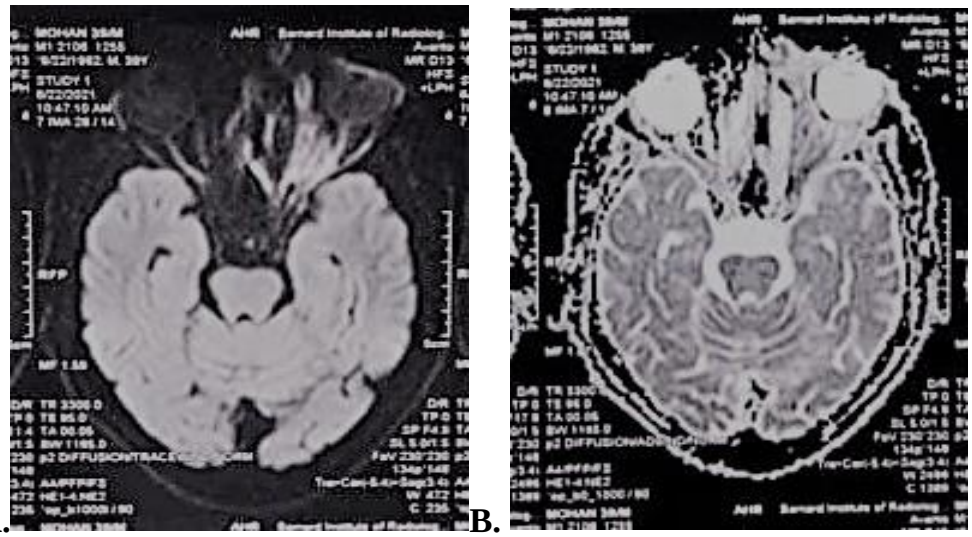


Fig. 29 MRI Contrast showing right orbital abscess.

Fig. Axial T2-weighted MR image of the orbits showing Right eye severe proptosis, periorbital edema, tenting of the posterior globe - ("Guitar pick" sign) and right ethmoid hyperintensity

2.7% of our study participants had evidence of optic nerve involvement in the form of restricted diffusion on MRI.

Desai et al.,2021; noted Optic nerve involvement could be discerned as heterogenous enhancement with diffusion restriction on MRI-DWI ADC sequencing [(97)].



A. **B.**

Fig. 30A. Diffusion Weighted Imaging showing heterogeneously enhancing left optic nerve and **B.** low apparent diffusion coefficient of left optic nerve - optic nerve infarctio

Fat stranding:

5.6% of our study population showed fat stranding in pre & retro antral soft tissue including infra temporal fossa.

Aribandi et al.,2007; recorded the involvement of orbital fat, extraocular muscles, peri antral fat as the harbinger of hyphal invasion [(98)].

Desai et al.,2021; stated Retro /pre/peri antral soft tissue inflammation could be identified as fat stranding on MRI [(97)].

Trigeminal and Meckel's cave involvement was seen in 1% of our study population on MRI.Desai et al.,2021; identified trigeminal and mandibular division of trigeminal nerve involvement on MRI in his study [(97)].

In our study most common manifestation of Intracranial extension was identified to be cavernous sinus thrombosis noted in 7.5% of participants. Other intracranial extension findings were intracerebral abscess in 4% of cases, infarct in 3% of cases, internal carotid artery involvement in 2.0% of cases with 2 cases having aneurysm, early cerebritis noted in 1% of cases, meningeal enhancement was noted in 0.7%and granuloma in 0.2% of total cases.

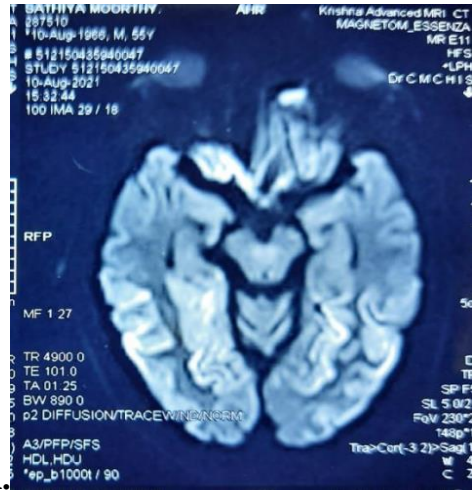
Aribandi et al.,2007; Observed that Leptomeningeal enhancement heralds the Intracranial extension of Mucormycosis which can progress to cerebritis, granuloma and cerebral abscess in advanced disease [(98)].

Desai et al.,2021; stated early Intracranial extension was discernible in the form of Dural enhancement along Basi frontal or Basi temporal region and in later stages as cerebritis, cavernous sinus & ICA involvement and abscess noted as hypo intensity on T1 & T2 and

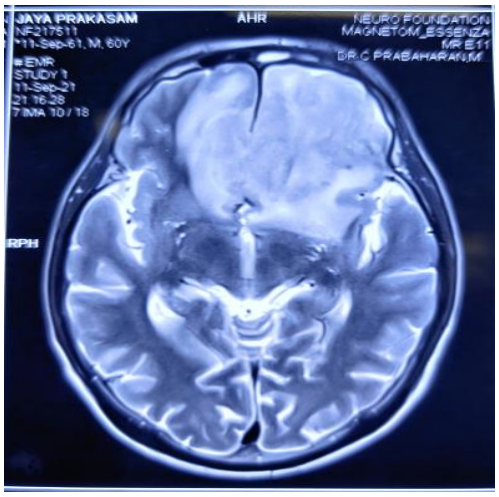
on contrast had peripheral rim enhancement [97]. CNS involvement in MRI was seen in the form of cavernous sinus thrombosis (53%), internal carotid artery occlusion (18%), temporal lobe abscess (12%), frontal lobe abscess (2.8%) and skull base osteomyelitis (7.1%) in a study by Sen et al., 2021;[13].

Involvement of cavernous sinus may have occured via pterygopalatine fossa, superior orbital fissure (1.2%), orbital apex (3.6%), mandibular nerve (1%) and direct extension via sphenoidal erosion (1.7%).

Among 23 intra cerebral abscesses 10 were found in temporal lobe, 10 in frontal lobe, 1 in both temporal and frontal lobe, one involving pons & cerebellar peduncle and 1 was intrasellar abscess.



A.



B.

Fig. 31 **A.**DWI sequence of MRI showing bilateral Parieto-occipital early cerebritis and **B.** Axial T2 weighted MR imaging showing cerebritis of bilateral frontal lobe

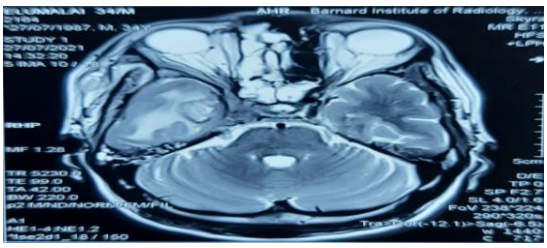
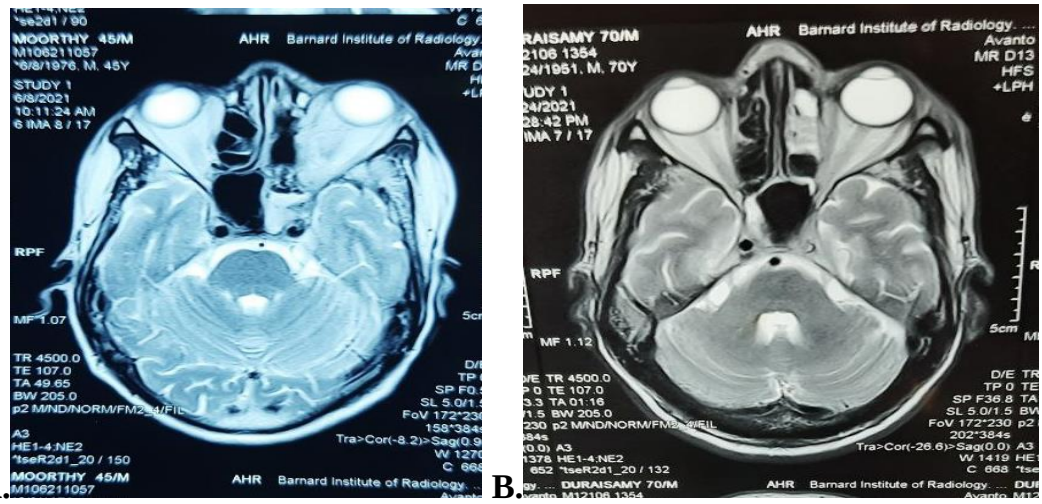


Fig. 32 Axial T2 weighted MR imaging showing granuloma in right temporal lobe



A. **B.**

Fig.A. Axial T2 weighted MRI shows left eye proptosis, cavernous sinus & Sphenoid involvement with patent internal carotid artery. **B.** T2 weighted MRI shows left cavernous sinus thrombosis with internal carotid artery thrombosis

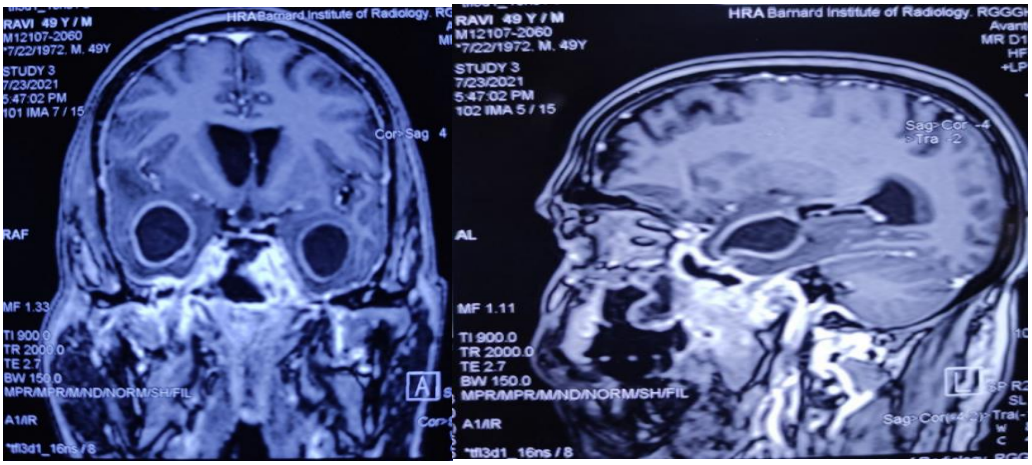


Fig. T1 contrast MRI Axial and Coronal cuts showing bilateral temporal lobe abscess. Sagittal image of the same patient showing right temporal abscess localizing to right greater wing of sphenoid area

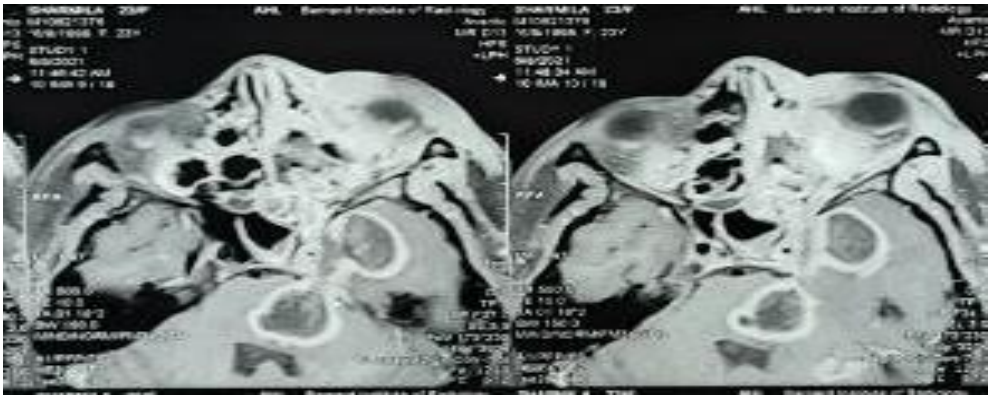


Fig. Axial MRI with contrast showing well encapsulated abscess with peripheral rim enhancement involving left temporal pole, Pons & cerebellar peduncle region abscess with left periorbital edema, proptosis and fat stranding.

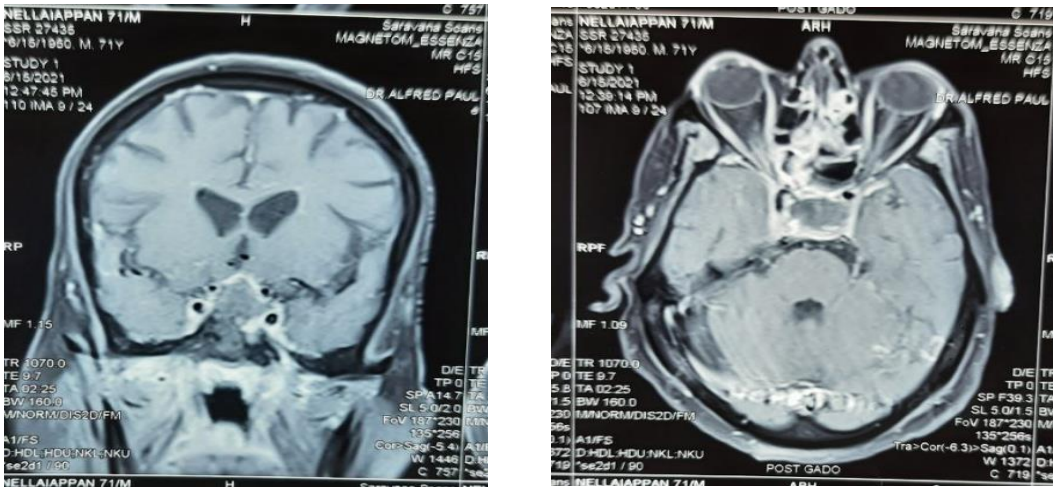


Fig. MRI coronal and axial cuts showing bilateral cavernous sinus involvement with sphenoid and intrasellar abscess

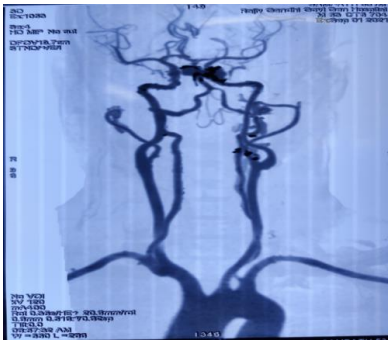


Fig. MRA showing aneurysm of left cavernous carotid artery

Olfactory sulcus involvement was noted in 1 case among our study population.

Desai et al.,2021; noticed Olfactory bulb involvement on MRI as heterogenous enhancement of the bulb [(97)].

Out of 162 participants with MRI, 156 individuals showed evidence of extra sinus invasion. Invasion was detected on MRI in 14 cases who turned out to be either non-invasive mucor or negative for fungal elements on HPE, which either indicates over reporting or sample turning negative due to delayed histo-pathological examination.

it is to be noted that in the 6 cases in our study with MRI that showed no evidence of invasion, 3 turned out to be negative for fungus while 3 showed angio and bone invasion on HPE.

STAGING:

Our study discovered that 63.6% of the participants developed severity of stage 3b or less and 28.3% had disease severity of stage 3c or above.

Sen et al., 2021; stated 49% of his study population had severity of stage 3b or less and 27% had stage 3c or above. [(13)]

MICROBIOLOGY

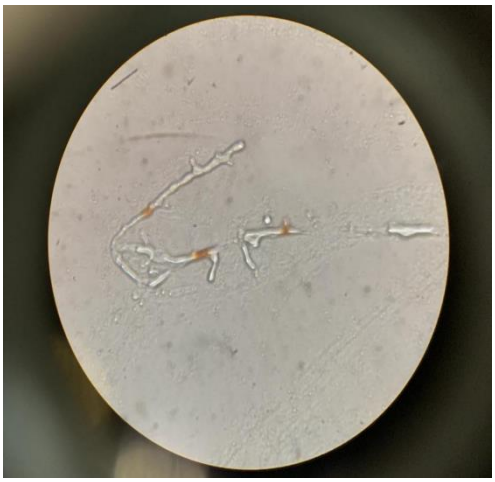
our study showed 69.3% KOH positivity for fungal hyphae, 74% HPE positivity and 16.4% culture positivity.

Mishra et al.,2021; described 90.6% **KOH** positivity, 84.3% showed fungal hyphae in **HPE** and 34.7% had fungus grown on **culture** [**(76)**].

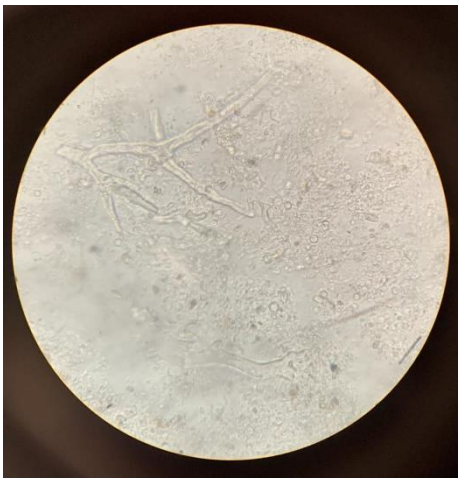
In the study by Sen et al., 2021; fungal hyphae were detected for 89% in KOH, 19% showed culture positivity and 39% were positive for mucor in histopathology [13].

In our study on comparison with HPE as gold standard, KOH had high sensitivity (72.2%) and positive predictive value (77.6%) highlighting the value of direct microscopy as a better screening tool, as it allows for rapid and prompt working diagnosis far before culture or histopathology.

Among the 403 positives for KOH in our study, 390(96.7%) showed Mucorales, 8 (2%) showed Aspergillus, 2 were mixed with 1 (0.2%) showing Candida and 1 (0.2%) showing Aspergillus in conjunction with Mucorales. KOH positivity may not definitely indicate an infection with Mucormycosis as it is a common commensal of the nasal cavity.



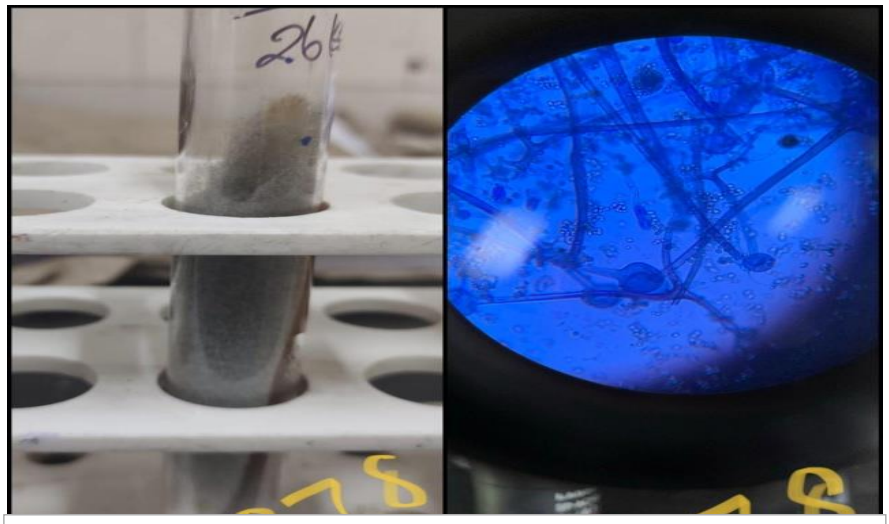
KOH mount demonstrating aseptate fungal hyphae



KOH mount demonstrating septate fungal hyphae with conidia

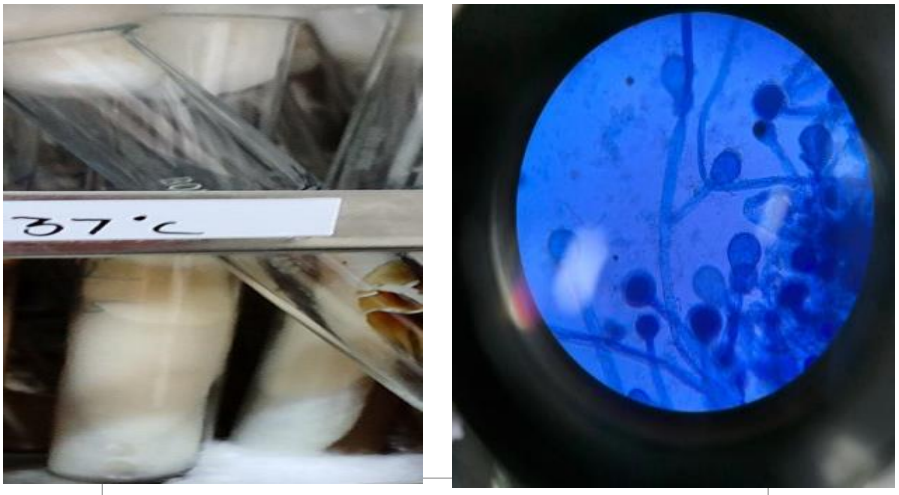
Among the 96 (16.4%) that showed fungal growth in culture in our study, Mucor was detected in 46 (48.0%), followed by Rhizopus in 38 (40.0%), Aspergillus in 10 (10%), Fusarium in 1(1.0%) and Syncephalastrum in 1 (1. %).

Fungi were difficult to grow in a medium due to high degree of contamination by bacteria and highly specific growth needs making it a less reliable diagnostic test.

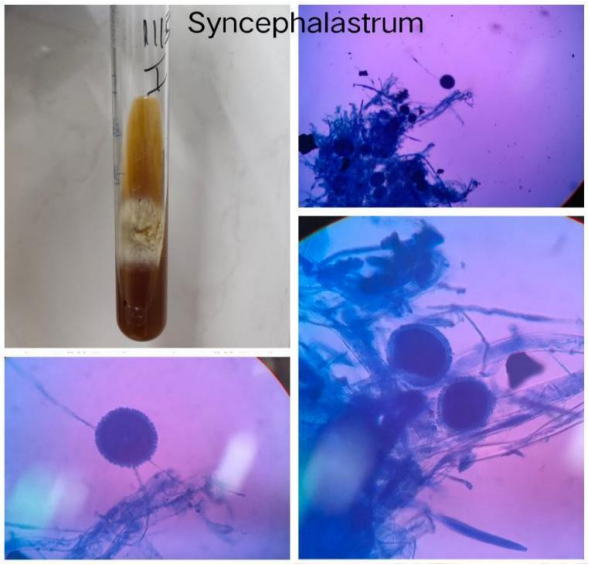


Rhizopus - culture & LPCB mount

Hematoxylin-eosin staining demonstrating Bone Invasion

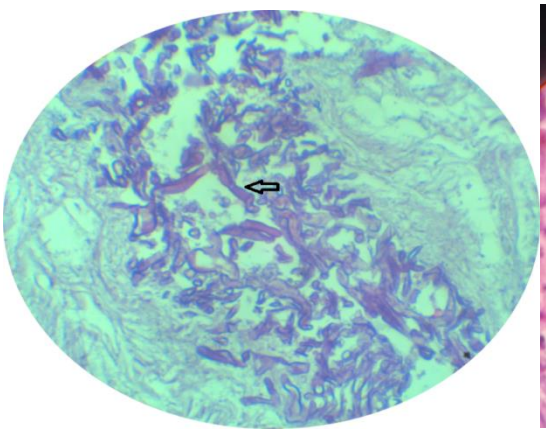


Mucor - culture & LPCB mount

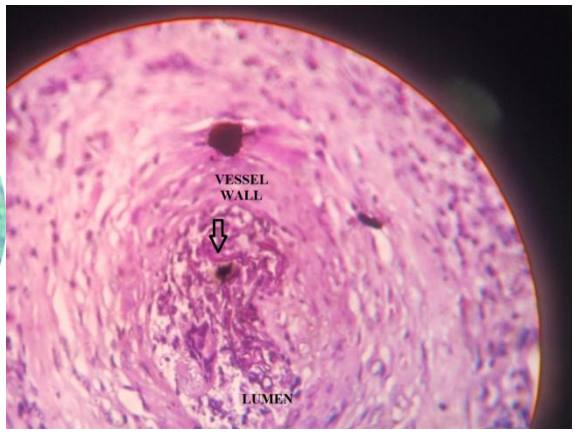


Syncephalastrum - culture & LPCB mount

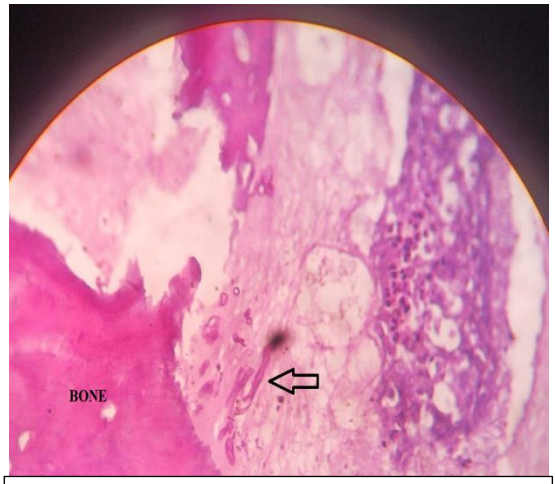
Among the 431 participants with proven with Mucormycosis on HPE, 96.3% (415) of the patients showed signs of invasion while the rest, 3.6%, had noninvasive Mucormycosis. Different types of invasions that were observed include angio invasion in 137 (31.7%), both angio and bony invasion in 256 (59.3%), bone invasion alone in 9 (2%), cartilage invasion in 2 (0.4%) and nerve invasion in 1 (0.2%).



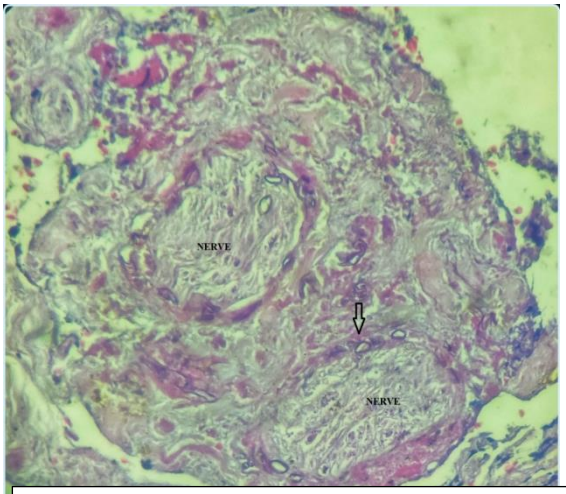
Hematoxylin-eosin staining showing fungal hyphae in tissue



Hematoxylin-eosin staining demonstrating angioinvasion



Hematoxylin-eosin staining demonstrating Bone Invasion



Hematoxylin-eosin staining demonstrating PeriNeural invasion

CONCLUSION:

- Covid Associated Mucormycosis is more common in males and in the fifth decade.
- The most common risk factors associated with Mucormycosis in our study are post Covid status, Diabetes Mellitus, hospitalization and steroid administration.
- This epidemic within a pandemic stresses the fact that vaccine against Covid-19 is an indispensable tool in fighting the pandemic & its associated afflictions and highlights the burning need for 100% vaccination against Covid-19.
- Covid Associated Mucormycosis was not associated with steam inhalation, industrial oxygen or zinc supplementation.
- The predominant symptoms seen in Covid associated Mucormycosis were discerned to be nasal obstruction, nasal discharge, facial swelling and facial pain.
- The earliest sign of orbital involvement is abduction restriction.
- Our study accentuates the ability of CT paranasal sinuses with orbital cuts in identifying early subtle fat stranding in extra sinus tissues and highlights the diagnostic value of CT in recognizing Mucormycosis at an early stage. Though MRI is preferred and is better at picking up soft tissue changes, CT should be favoured due to its limited cost and easy accessibility with MRI being reserved for cases suspected to have advanced stage disease to promptly identify the same.
- Direct microscopy is a better screening tool for early diagnosis and initiation of treatment in Covid Associated Mucormycosis.

- with the limitations of the diagnostic tests being evident, a need for a more elaborate diagnostic criteria for the early detection and initiation of treatment in Mucormycosis is imperative and the following criteria is flexible and reliable:

Possible:

Equivocal Nasal findings+ Mycology positive + Radiology equivocal

(or)

Nasal Endoscopy positive+ Mycology negative + Radiology positive/equivocal

Probable:

Doubtful Nasal findings + Mycology positive + Radiology positive

Definitive:

Nasal Endoscopy positive +Mycology positive+ Radiology positive

Certain:

Definitive + Histopathology positive

Probable + Histopathology positive

Possible + Histopathology positive

LIMITATIONS

1. our study was hospital based, not a community-based study
2. The possibility of recall bias among the study population while data collection
3. With many patients having treated for Covid elsewhere, the proper details regarding the Covid management couldn’t be collected satisfactorily