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# A RETROSPECTIVE STUDY OF ETIOLOGICAL AGENTS OF FUNGAL RHINOSINUSITIS FROM A TERTIARY CARE HOSPITAL IN HIMACHAL PRADESH.

Clinical Microbiology

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## ABSTRACT

**Background and objective:** Rhinosinusitis, clinically is co-existence of both sinusitis and rhinitis which cannot be differentiated physiologically and pathophysiologically from one another and are diagnostic terminologies related to clinical features of effected para-nasal sinuses and the nose. Fungal rhinosinusitis (FRS) can be acute or chronic depending upon the duration of symptoms as well as invasive and non-invasive form based on histopathological evidence. Fungi are common cause of rhinosinusitis and incidence is increasing day by day. One year retrospective study w.e.f. August 2022 to July 2023 in a tertiary care hospital was done to evaluate the fungal profile of clinically suspected patients of FRS. **Methodology:** A total of 44 samples were processed during the study period and were subjected to direct microscopic examination by KOH mount and fungal culture. Samples includes nasal crusts, nasal scrapings , tissue biopsy samples following FESS (Functional endoscopic sinus surgery) and pus from sinuses. **Result:** In 44 samples , fungal elements were seen in 7 (15.91% ) and no fungal elements were seen in 37 ( 84.09%) on KOH mount. Fungal growth was obtained in 6 ( 13.64%) and no fungal growth obtained in 38 ( 86.36% ) samples on fungal culture. *Rhizopus* was the most common isolate ( 42.86%) followed by *Aspergillus* ( 28.58%). Early diagnosis and treatment of FRS is important to reduce the morbidity and mortality associated with angio-invasive nature of common etiological fungal agents.

## KEYWORDS

FRS (fungal rhinosinusitis), FESS, KOH, LPCB stain, SDA,

## INTRODUCTION:

Rhinosinusitis in inflammation of nasal and paranasal sinus mucosa associated with mucosal alterations[1]. Rhinosinusitis can be acute which include disease lasting for 7 days to < 4 weeks ; sub-acute with duration of 4-12 weeks; recurrent acute rhinosinusitis that has ≥4 episodes per year and chronic rhinosinusitis (CRS) is a disease lasting for >12 weeks[2,3]. Fungal rhinosinusitis (FRS) can be acute and chronic depending upon the duration of disease. FRS is one of the important health care problem and its incidence is increasing over the past three decades[4]. The improved techniques in Mycology, Histopathology and Radiology has led to improved disease detection rate and also alteration in normal microbiota of nasal and paranasal sinuses due to indiscriminate use of antimicrobial agents as well as increase in number of immunocompromised individuals has led to increases cases of FRS[5]. FRS can occur in both immunocompetent and immunocompromised individuals with chronic, indolent progression in former and rapidly progressive and fulminant in latter[6]. North India has been identified as endemic zone of paranasal mucoses[4].

Fungal ball, allergic fungal rhinosinusitis (AFRS), acute invasive FRS, granulomatous invasive FRS, chronic invasive FRS are types of FRS. *Aspergillus , Mucor, Rhizopus* spp. Are the commonest fungi causing FRS . *Bipolaris, Curvularia, Alternaria, Candida, Fusarium* spp. etc rare causative agents[6,7,8].

## AIMS AND OBJECTIVE:

A retrospective study of the fungal etiological agents among clinically suspected patients of fungal rhinosinusitis over a period of one year.

## METHODOLOGY:

This retrospective study was conducted at Dr. Rajendra Prasad Government Medical College Kangra at Tanda, Himachal Pradesh w.e.f. 1st August 2022 to 31st July 2023. A total of 44 samples were received in Mycology laboratory from E.N.T. department with clinical suspicion of fungal rhinosinusitis and were processed as per standard operating procedures. The specimens include nasal crusts, nasal scrapings, tissue biopsy following functional endoscopic sinus surgery (FESS) , pus from sinus in sterile universal container with or without normal saline (0.85%).

### Direct microscopic examination :

Slide KOH (Potassium hydroxide) (10%) mount [9] was done for all samples received in laboratory. Tube KOH (20-40%) was done in case of improperly dissolved samples. Fungal elements were observed and reported accordingly.

Fungal culture [10] on two sets of Sabouraud's dextrose agar (SDA) with or without antibiotics at 25°C and 37°C was done which were observed for growth for 4weeks. If no growth was observed at the end of 4 weeks, the cultures were considered negative for fungal growth. The growth positive cultures were observed for growth chara cteristics, texture, surface, color of growth and pigment production.

Gram's stain was done for smooth , moist, pasty colonies and for Gram positive budding yeast cells, germ tube test, Dalmau plate culture was done and also sub-cultured on candida CHROMagar for identification of species. Lactophenol-cotton blue (LPCB) stain preparation[11,12] , tease mount preparation using LPCB stain and slide culture preparation[12] was done to study the morphological details and sporulation of molds. All observations made on direct microscopic examination, culture characteristics, microscopic findings observed on further processing of fungal culture were used to identify the fungal species.

## RESULTS:

Total samples received were 44 , out of which 36 were males and 08 were females.

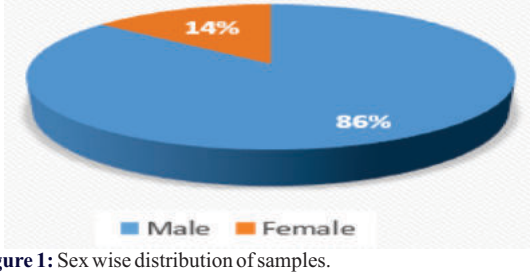


**Figure 1:** Sex wise distribution of samples.

**Table 1:** Age wise distribution of samples and fungal positivity of samples.

| S.No: | Age group (yrs) | No. Of samples | KOH positive | Culture positive |
|---|---|---|---|---|
| 1. | 1-18 | 01 | 00 | 00 |

| | | | | |
|---|---|---|---|---|
| 2. | 19-30 | 05 | 01 | 00 |
| 3. | 31-40 | 05 | 00 | 00 |
| 4. | 41-50 | 11 | 02 | 03 |
| 5. | 51-60 | 10 | 03 | 01 |
| 6. | > 60 | 12 | 01 | 02 |
| Total | | 44 | 07 | 06 |

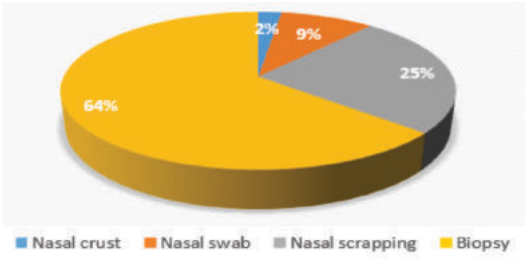


**Figure 2:** Distribution of sample type received.

**Table 2:** Direct Microscopy- KOH mount examination.

| S.No: | Fungal elements | Number | Percentage (%) |
|---|---|---|---|
| 1. | KOH positive | 07 | 15.91 |
| 2. | KOH negative | 37 | 84.09 |
| Total | | 44 | 100 |

**Table 3:** Fungal culture report of samples.

| S.No: | Fungal culture | Number | Percentage (%) |
|---|---|---|---|
| 1. | Growth positive | 06 | 13.64 |
| 2. | Growth negative | 38 | 86.36 |
| Total | | 44 | 100 |

**Table 4 :** 10% KOH Mount and Fungal cultures

| KOH Mount | Number | Fungal growth obtained | No fungal growth |
|---|---|---|---|
| Positive | 07 | 03 | 04 |
| Negative | 37 | 03 | 34 |
| Total | 44 | 06 | 38 |

A total of 10 (22.72%) were fungal positive including both KOH positivity and fungal culture growth positive and rest 34 samples were negative.

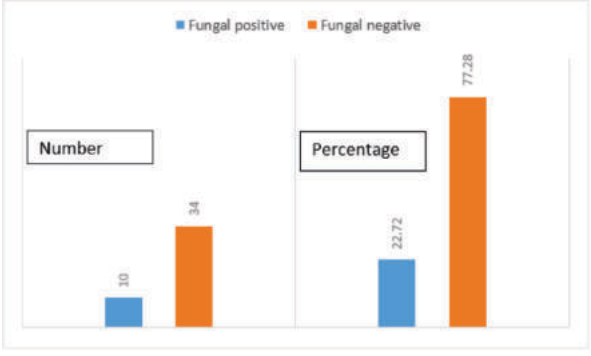


**Figure 3**: Fungal positivity of samples.

**Table 5:** Fungal isolates obtained .

| S.No: | Fungal isolate | Number | Percentage (%) |
|---|---|---|---|
| 1. | Rhizopus spp. | 03 | 42.86 |
| 2. | Aspergillus spp. | 02 | 28.58 |
| 3. | Curvularia spp. | 01 | 14.28 |
| 4. | Alternaria spp. | 01 | 14.28 |
| Total | | 07 | 100 |

## DISCUSSION:

The present study was done to know the common etiological agents of FRS among clinically suspected patients from all age groups. Out of total 44 samples received and processed in mycology laboratory KOH positive samples were 7 (15.91%) while fungal culture growth positive were 6 (13.64%). KOH positivity is more than culture positivity and is in concordance with studies of Virdi S.L. *et al*[13] and Kulkarni S.S. *et al* [14]. Overall fungal positivity including direct microscopy and culture were seen in 10 (22.72%) samples which is similar to study by Shivani *et al* [15]. Both KOH positive and fungal culture positive samples were 3 (30%) out of 10 fungal positive samples while 4 (40%) were only KOH positive and culture negative. Rest 3 (30%) were KOH negative but fungal culture positive. A total of 7 fungal isolates were obtained on fungal culture. Most common isolate was *Rhizopus* spp. ( 42.86%) followed by *Aspergillus* spp. (28.58%) and other isolates were *Curvularia* spp.(14.28) and *Alternaria* spp. (14.28%) .

## CONCLUSION:

- Fungal rhinosinusitis is a serious disease with significant morbidity and even mortality due to invasive nature and more so in diabetic and immunocompromised patients.
- Angioinvasive nature of *Rhizopus* spp. ,*Aspergillus* spp. and *Mucor* spp. can lead to more severe disease and complications like orbital and intra-cranial manifestations. Prompt diagnosis and early initiation of antifungal treatment is essential to reduce both morbidity and complications.
- Small sample size was not sufficient for significant results and other methods like histopathological co-relation and molecular methods should be used for diagnosis of FRS.
- Fungal culture directed therapy is gold standard for the management of FRS.

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**Conflict of Interest / Competing interests:**

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