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Original Article

Ear, Nose and Throat Section

Diagnostic Accuracy of Skin Prick Test in Invasive Fungal Sinusitis: A Cross-sectional Study

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ABSTRACT

Introduction: Invasive Fungal Sinusitis (IFS) is a life-threatening infection with a rapid progression. Early diagnosis is crucial for optimal patient outcomes. Skin Prick Testing (SPT) is a simple, non invasive test that could potentially aid in early detection.

Aim: To assess the diagnostic accuracy of SPT in identifying IFS caused by *Mucor*, *Aspergillus*, *Rhizopus* and *Fusarium*.

Materials and Methods: A cross-sectional study was conducted at Department of Ear, Nose and Throat (ENT), KEM Hospital, Pune, Maharashtra, India, from August 2017 to April 2019. Patients with clinically and radiologically suspected IFS were evaluated and the diagnosis was confirmed by histopathological examination of sinonasal tissue, which was considered the gold standard. The primary outcome measured was the diagnostic utility of SPT by comparing SPT results with histopathological findings. Statistical analysis included calculation of sensitivity, specificity, Positive Predictive Value (PPV) and Negative Predictive Value (NPV), with categorical variables analysed using the Chi-square test or Fisher's-exact test, as appropriate.

Results: A total of 25 patients with IFS were enrolled, with a mean age of 63.3±10.1 years. SPT revealed positive results in 13/25 (52%) of patients for *Mucor*, 9/25 (36%) for *Aspergillus*, 2/25 (8%) for *Fusarium* and 1/25 (4%) for *Rhizopus*. SPT for *Mucor* demonstrated high sensitivity (81.2%) and specificity (100%), while SPT for *Aspergillus* showed perfect diagnostic accuracy (100% sensitivity, specificity, PPV, NPV and accuracy), whereas SPT for *Fusarium* and *Rhizopus* sensitivity could not be calculated and 0% PPV despite high specificity (92% and 96%, respectively), limiting their diagnostic value.

Conclusion: The SPT demonstrated excellent diagnostic accuracy for *Aspergillus* and good accuracy for *Mucor*, whereas it had limited value for *Fusarium* and *Rhizopus*.

Keywords: Allergens, Aspergillosis, Invasive fungal infections, *Mucor*, Mucormycosis, Skin tests

INTRODUCTION

Fungal infections of the sinonasal tract are increasingly recognised, particularly among immunocompromised individuals [1,2]. Fungal sinusitis can be classified as non invasive, where fungi colonise the sinuses without tissue invasion and invasive, characterised by fungal penetration into the mucosa, bone, or surrounding tissues, often leading to severe morbidity and mortality [2]. IFS is a rapidly progressive and potentially fatal condition, commonly caused by organisms such as *Mucor*, *Aspergillus*, *Rhizopus* and *Fusarium* [2]. Delayed diagnosis is a major contributor to poor clinical outcomes, as conventional diagnostic methods often rely on invasive procedures and time-consuming microbiological or histopathological confirmation [2].

The authors hypothesised that sensitisation to fungal antigens detected by SPT may be associated with the presence of IFS and could assist in early identification of high-risk individuals. The present study was therefore conducted to evaluate the diagnostic accuracy of SPT in detecting IFS caused by *Mucor*, *Aspergillus*, *Rhizopus* and *Fusarium*.

MATERIALS AND METHODS

The present study was a cross-sectional study conducted at Department of ENT, KEM Hospital, Pune, Maharashtra, India, a 600-bed tertiary care hospital, from August 2017 to April 2019. The study protocol was approved by the Institutional Ethics Committee (765/2017). All procedures were conducted in accordance with the declaration of Helsinki.

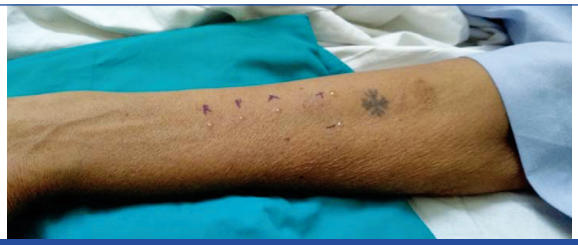
Consecutive patients meeting the inclusion criteria during the study period were enrolled using a convenience sampling technique.

Inclusion and Exclusion criteria: All patients with clinical suspicion of IFS based on symptoms (facial pain, nasal obstruction, or discharge), radiological findings {Computed Tomograhy (CT)/ Magnetic Resonance Imaging (MRI) suggestive of sinus invasion} and intraoperative features were evaluated. The diagnosis of IFS was confirmed by histopathological examination of sinonasal tissue specimens, which served as the gold standard. All patients with histopathologically proven IFS were included in the study. Patients with severe generalised skin disease, a history of anaphylaxis, or those who declined to participate were excluded.

Study Procedure

Detailed medical history, including co-morbidities, medications and previous treatments, was recorded and physical examination findings were documented.

Skin Prick Testing (SPT): The SPT was performed using a standardised technique on the volar surface of the forearm [Table/Fig-1]. A panel of fungal allergens (*Mucor*, *Aspergillus*, *Rhizopus* and *Fusarium*) was used, along with histamine as a positive control and saline as a negative control. Wheal and flare reactions were measured 20 minutes post-prick. A wheal diameter ≥3 mm than the negative control was considered a positive reaction. This methodology follows established guidelines for SPT in allergic and fungal sensitisation studies [3].



[Table/Fig-1]: Performing skin prick test.

Comparison with Histopathology: All patients underwent histopathological examination of sinonasal tissue obtained during diagnostic or therapeutic procedures. The SPT was performed prior to tissue sampling. SPT results were compared with histopathology, which served as the gold standard for diagnosis.

STATISTICAL ANALYSIS

Data were analysed using Statistical Package for the Social Sciences (SPSS) version 21.0 (IBM Corp., Armonk, NY, USA). Categorical variables were summarised as frequencies and percentages. Continuous variables were presented as mean ± Standard Deviation (SD) with range. The diagnostic performance of SPT for each fungal allergen (*Mucor*, *Aspergillus*, *Fusarium* and *Rhizopus*) was evaluated against histopathology as the gold standard. Sensitivity, specificity, PPV, NPV and overall accuracy were calculated using standard formulas. The agreement between SPT and histopathology was assessed using Cohen's kappa coefficient (κ), with values interpreted as follows: <0.20, poor; 0.21-0.40, fair; 0.41-0.60, moderate; 0.61-0.80, good; and 0.81-1.00, very good agreement. For categorical comparisons, the Chi-square test or Fisher's-exact test (for expected cell counts <5) was applied as appropriate. All tests were two-tailed and a p-value <0.05 was considered statistically significant.

RESULTS

A total of 25 patients with IFS were enrolled in the present study. The majority were males 16/25 (64%) and 9/25 (36%) were females, with a male-to-female ratio of 1.78:1. The mean age was 63.3±10.1 years (range 42-89 years). A total of 10 patients (40%) had a history of allergy. A total of 17 patients (68%) had diabetes mellitus, 5 patients (20%) had haematological malignancies and 3 patients (12%) were immunocompromised due to prolonged corticosteroid use. The most common presenting symptoms were facial pain in 20 patients (80%), nasal obstruction in 18 patients (72%) and purulent nasal discharge in 15 patients (60%) [Table/Fig-2].

Variables		n (%)
Demographics	Male	16 (64%)
	Female	9 (36%)
	Male: Female ratio	1.78 : 1
	Age (years), mean±SD	63.3±10.1
	Age range (years)	42-89
Medical history/ co-morbidities	History of allergy	10 (40%)
	Diabetes mellitus	17 (68%)
	Haematological malignancy	5 (20%)
	Prolonged corticosteroid use	3 (12%)
Presenting symptoms	Facial Pain	20 (80%)
	Nasal obstruction	18 (72%)
	Purulent nasal discharge	15 (60%)

[Table/Fig-2]: Demographic characteristics, co-morbidities and clinical presentation of patients with Invasive Fungal Sinusitis (IFS) (N=25).

Skin Prick Test Results

Mucor: 13/25 (52%) patients had a positive SPT for *Mucor* [Table/Fig-3]. *Aspergillus*: 9/25 (36%) patients had a positive SPT for *Aspergillus* [Table/Fig-4]. *Fusarium*: 2/25 (8%) patients had a positive SPT for *Fusarium* [Table/Fig-5] *Rhizopus*: 1/25 (4%) patients had a positive SPT for *Rhizopus* [Table/Fig-6].

Mucor by skin prick test	Positive n (%)	Negative n (%)	Kappa-value	p-value
Positive	13 (81.3)	0	0.757	0.001***
Negative	3 (18.7)	9 (100.0)		
Total	16 (100.0)	9 (100.0)		

[Table/Fig-3]: Distribution of detection of *Mucor* fungal sinonasal infection by skin prick test and by histopathology.
*p-value by (Fisher's-exact probability test)

Aspergillus by skin prick test	Positive n (%)	Negative n (%)	Kappa-value	p-value
Positive	9 (100.0)	0	1.000	0.001***
Negative	0	16 (100.0)		
Total	9 (100.0)	16 (100.0)		

[Table/Fig-4]: Distribution of detection of *Aspergillus* fungal sinonasal infection by skin prick test and by histopathology.
*p-value by Fisher's-exact probability test).

Fusarium by skin prick test	Positive n (%)	Negative n (%)	Kappa-value	p-value
Positive	0	2 (8.0)	--	--
Negative	0	23 (92.0)		
Total	0	25 (100.0)		

[Table/Fig-5]: Distribution of detection of *Fusarium* fungal sinonasal infection by skin prick test and by histopathology.
p-value and Kappa-value was not possible due to insufficient data.

Rhizopus by skin prick test	Positive n (%)	Negative n (%)	Kappa-value	p-value
Positive	0	1 (4.0)	--	--
Negative	0	24 (96.0)		
Total	0	25 (100.0)		

[Table/Fig-6]: Distribution of detection of *Rhizopus* fungal sinonasal infection by skin prick test and by histopathology.
p-value and Kappa-value was not possible due to insufficient data

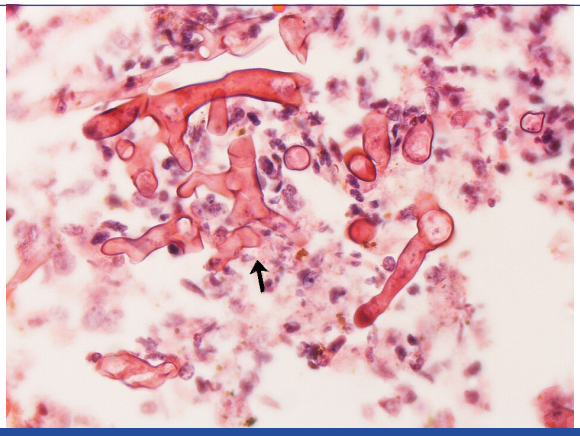
No systemic or local adverse reactions were observed during or after the SPT procedure and all tests were well-tolerated by the patients.

Comparison with Histopathology

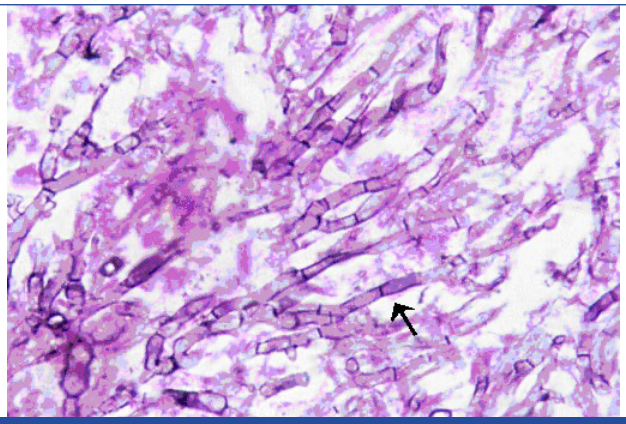
Mucor: SPT for *Mucor* demonstrated a sensitivity of 81.2%, specificity of 100%, PPV of 100%, NPV of 75% and accuracy of 88%. *Aspergillus*: SPT for *Aspergillus* showed perfect sensitivity, specificity, PPV, NPV and accuracy (100%). *Fusarium* and *Rhizopus*: SPT for *Fusarium* and *Rhizopus* had limited diagnostic accuracy, with low sensitivity and specificity [Table/Fig-7]. Histopathological images of the *mucor* and *aspergillus* are shown in [Table/Fig-8,9].

Detection	Sensitivity	Specificity	Positive Predictive Value (PPV)	Negative Predictive Value (NPV)	Accuracy
Mucor	81.2	100.0	100.0	75.0	88.0
Aspergillus	100.0	100.0	100.0	100.0	100.0
Fusarium	--	92.0	0.0	100.0	92.0
Rhizopus	--	96.0	0.0	100.0	96.0

[Table/Fig-7]: Detection efficacy indices of skin prick test against histopathology as a gold standard for various types of detection.



[Table/Fig-8]: Histopathological appearance of *mucor* (1000x, H&E).



[Table/Fig-9]: Histopathological appearance of *Aspergillus fumigatus* (400x, H&E).

DISCUSSION

In the present cross-sectional study of 25 patients with histopathologically confirmed IFS, SPT demonstrated excellent diagnostic performance for *Aspergillus*, with 100% sensitivity and specificity and good performance for *Mucor*, with 81.2% sensitivity and 100% specificity.

Early identification of individuals at risk of IFS is therefore crucial for timely intervention and improved survival. SPT is a well-established, rapid, non invasive and cost-effective method used for detecting Immunoglobulin E (IgE)-mediated hypersensitivity to specific allergens [4]. Epidemiological studies suggest that IFS predominantly affects immunocompromised patients, including those with diabetes, haematological malignancies, or prolonged corticosteroid use, with a rising incidence reported worldwide [5-7].

The study has several strengths, including the use of histopathology as the gold standard for all patients, a comprehensive SPT panel covering common causative fungi and a standardised methodology with blinded assessment, minimising measurement bias. Clinically, SPT offers a rapid, non invasive screening tool that may facilitate early identification of high-risk patients, particularly those with diabetes mellitus or other immunocompromised states. Positive SPT results for *Aspergillus* or *Mucor* can prompt earlier imaging, intervention and antifungal therapy, potentially improve outcomes and reducing the need for invasive procedures when histopathology or culture is delayed or unavailable.

Limitation(s)

The small sample size limits the generalisability of the findings and the cross-sectional design prevents assessment of the longitudinal predictive value of SPT. Additionally, SPT showed limited utility for *Fusarium* and *Rhizopus*, which may be influenced by low environmental exposure, allergenicity, or regional prevalence. The specificity of SPT may also be affected by prior fungal exposure or immunological status.

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CONCLUSION(S)

The SPT demonstrates excellent diagnostic accuracy for *Aspergillus* and good accuracy for *Mucor*, suggesting its value as an early, non invasive tool for identifying patients at risk of IFS. Further large-scale, prospective studies are warranted to validate these findings and clarify the clinical utility of SPT for less common fungal pathogens such as *Fusarium* and *Rhizopus*.

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