

# Comparative Analysis of Antifungal Susceptibility in Trichophyton Species Using Broth Microdilution Assay: A Cross-Sectional Study

Anshu Singh <sup>1</sup>, Amber Prasad <sup>1</sup>, Gagandeep Singh <sup>2</sup>, Neerita Hazarika <sup>3</sup>, Pratima Gupta <sup>4</sup>, Neelam Kaistha <sup>1</sup>

Review began 12/24/2024

Review ended 12/30/2024

Published 01/07/2025

© Copyright 2025

Singh et al. This is an open access article distributed under the terms of the Creative Commons Attribution License CC-BY 4.0., which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

DOI: 10.7759/cureus.77113

<sup>1</sup>. Microbiology, All India Institute of Medical Sciences, Rishikesh, Rishikesh, IND <sup>2</sup>. Microbiology, All India Institute of Medical Sciences, New Delhi, New Delhi, IND <sup>3</sup>. Dermatology, Venereology, and Leprosy, All India Institute of Medical Sciences, Guwahati, Guwahati, IND <sup>4</sup>. Microbiology, All India Institute of Medical Sciences, Deoghar, Deoghar, IND

**Corresponding author:** Anshu Singh, anshusingh05july@gmail.com

## Abstract

### Background

Dermatophyte infections, predominantly caused by *Trichophyton* species, are a global health concern, particularly in tropical regions. The rising prevalence of resistant strains, due to the misuse of antifungal agents, necessitates robust antifungal susceptibility testing. This study aimed to evaluate antifungal susceptibility patterns of clinical isolates using the broth microdilution (BMD) assay to guide treatment strategies effectively.

### Methods

A cross-sectional study was conducted at the All India Institute of Medical Sciences (AIIMS), Rishikesh, for over two years, including 184 clinical samples from patients with recalcitrant dermatophytosis. Samples were collected aseptically, and antifungal susceptibility testing was performed using the BMD assay. Minimum inhibitory concentration (MIC) values were analyzed for five drugs: amphotericin, terbinafine, griseofulvin, itraconazole, and fluconazole. Associations with patient demographics, potassium hydroxide (KOH) positivity, and culture outcomes were statistically assessed.

### Results

Amphotericin showed the lowest mean MIC (0.57 µg/mL), while fluconazole exhibited the highest (7.70 µg/mL). Amphotericin and terbinafine achieved 100% inhibition, while other drugs showed 80% inhibition. Gender had no significant effect on MIC values. KOH positivity and culture outcomes significantly influenced MICs, with species-specific patterns observed. For instance, *T. mentagrophytes* and *T. indotineae* were linked to higher MICs for fluconazole and griseofulvin.

### Conclusion

The study underscores the need for species-specific antifungal susceptibility testing to monitor resistance trends. Results focused on unexpected resistance to typically effective drugs like terbinafine or fluconazole. Routine testing and targeted stewardship are essential to improve therapeutic outcomes for dermatophytosis.

**Categories:** Other, Pathology, Dermatology

**Keywords:** amphotericin and terbinafine, antifungal susceptibility, culture outcomes, dermatophyte, koh positivity, mics

## Introduction

Dermatophyte infections, predominantly caused by *Trichophyton* species, are a growing global health concern, especially in tropical and subtropical regions where warm and humid conditions favor fungal growth and transmission. These infections, commonly presenting as tinea, affect keratinized tissues such as the skin, hair, and nails, and can result in chronic, recurrent conditions and significant morbidity if left untreated or managed ineffectively [1]. The increasing prevalence of these infections, compounded by the widespread misuse and overuse of antifungal agents, has contributed to the emergence of resistant *Trichophyton* strains, posing challenges for effective clinical management [2]. The declining efficacy of frontline antifungal agents, such as azoles and allylamines, has been documented globally, with resistance rates varying across regions and species [3]. These trends emphasize the challenges posed by the rise in antifungal resistance due to overprescription or misuse of treatments and the urgent need for robust antifungal susceptibility testing to inform appropriate treatment strategies. The broth microdilution (BMD) assay, endorsed by the Clinical and Laboratory Standards Institute (CLSI), is a gold standard method for determining minimum inhibitory concentrations (MICs) of antifungal drugs [4]. Its quantitative and

### How to cite this article

Singh A, Prasad A, Singh G, et al. (January 07, 2025) Comparative Analysis of Antifungal Susceptibility in Trichophyton Species Using Broth Microdilution Assay: A Cross-Sectional Study. Cureus 17(1): e77113. DOI 10.7759/cureus.77113

reproducible results are crucial for identifying emerging resistance patterns and guiding therapeutic interventions. Recent studies have highlighted significant variability in antifungal susceptibility among *Trichophyton* species, with notable resistance to terbinafine, a commonly prescribed antifungal, in certain geographic regions [5,6]. These findings underscore the importance of species-specific monitoring and susceptibility profiling. This cross-sectional observational study aims to conduct a comparative analysis of antifungal susceptibility in clinical isolates of *Trichophyton* species using the BMD assay. The study's findings will contribute to a deeper understanding of resistance patterns and support evidence-based antifungal stewardship to combat dermatophytosis effectively. The primary objective of this study is to evaluate the antifungal susceptibility and resistance profiles of *Trichophyton* species isolated from dermatophyte infections in order to inform more targeted treatment strategies.

## Materials And Methods

It was a cross-sectional study performed at the All India Institute of Medical Sciences (AIIMS), Rishikesh, over two years. This study started from 22 August 2022 to 7 September 2024.

### Sample size

The study is based on a population size (N) of 660, which is derived from assuming 330 samples per year over a study duration of two years. The hypothesized percentage frequency of the outcome factor in the population (p) is 83%, with an absolute precision of 5%. The design effect (DEFF) for cluster surveys is set at 1. For a 95% confidence level, the required sample size is 164. Assuming 10% sample wastage, estimated sample size was 183 [7].

### Inclusion criteria

All samples (nail, skin, hair) of clinical cases of recalcitrant dermatophytosis were obtained from patients who attended the dermatology department of AIIMS Rishikesh.

### Exclusion criteria

Patients with naive infections.

### Sample collection

Aseptic conditions were used to collect the samples. Before gathering a clinical specimen, preferably from the expanding edge of lesions, the affected location was cleaned with alcohol.

Skin scrapings were gathered from center to edge, crossing boundaries with the use of a sterile scalpel blade held at an angle of 90° to the skin surface. When hair involvement occurred, we plucked the affected hair instead of clipping it. Depending on the type of nail infection, nail clippings were taken from the relevant location.

The material was gathered into folded squares of black paper, which allowed the specimen to dry, decreased bacterial contamination, and created conditions that allowed the specimens to be kept for extended periods of time without significantly compromising the fungal agents' vitality.

### Direct microscopy

Potassium hydroxide (KOH) wet mount: The cement substance that keeps keratinized cells together dissolves, and protein waste is softened and digested by the aqueous potassium hydroxide (KOH). On a microscopic slide, the specimen is submerged in a 10% KOH drop and covered with a cover slip. In clinical specimens, the fungal components become easily visible and rather transparent.

Depending on the type of clinical material, the KOH concentration can be raised; for solid specimens, it can reach 40%, while for soft specimens, it can reach 10%.

### Culture

Regardless of the results of a direct examination, the clinical specimens ought to be inoculated on fungal culture media. On Sabouraud dextrose agar (SDA) supplemented with antibiotics and cycloheximide, dermatophytes can grow readily. The media are incubated at three different temperatures: 25°C, 30°C, and 37°C.

It often takes between ten days and three weeks for the growth to occur. Colony morphology, lactophenol cotton blue (LPCB) mount, and slide culture were performed to examine the morphology of micro- and macroconidia following growth.

### Antifungal susceptibility testing

The Clinical and Laboratory Standards Institute (CLSI) criteria, document M38-A2, for filamentous fungi, were followed in the performance of the in vitro antifungal susceptibility assay. The antifungal agent stock solutions were made for the medications itraconazole (Himedia), fluconazole (Himedia), griseofulvin (Sigma Aldrich), terbinafine (Sigma Aldrich), and amphotericin-B (Himedia). Antifungal susceptibility testing was conducted using Rose Parker Memorial Institute-1640 (RPMI-1640) as a growth medium (pH 7.0 ± 0.1).

#### *Weighing Antifungal Powders*

Assay all antifungal agents for standard units of activity. The assay units can differ widely from the actual weight of the powder and often differ within a drug production lot. Thus, a laboratory must standardize its antifungal solutions based on assays of the lots of antifungal powders used.

Use either of the following formulas to determine the amount of powder or diluent needed for a standard solution:

$$\text{Weight (mg)} = \text{Volume (mL)} \times \text{Concentration (\mu g/mL)} \div \text{Potency (\mu g/mg)}$$

or

$$\text{Volume (mL)} = \text{Weight (mg)} \times \text{Potency (\mu g/mg)} \div \text{Concentration (\mu g/mL)}$$

The antifungal powder should be weighed on an analytical balance that has been calibrated by approved reference weights from a national metrology organization. Usually, it is advisable to accurately weigh a portion of the antifungal agent in excess of that required and to calculate the volume of diluent needed to obtain the concentration desired.

Dermatophytic strains were cultivated on SDA for seven to 14 days to create stock inocula. Colonies were then submerged in 5 ml of sterile water, and the suspension was created by gently probing the surface with the tip of a sterile device. After transforming the suspended mixture in a sterile tube and letting it settle for fifteen minutes, the upper homogenous suspension was employed for additional testing.

The inoculum concentration was adjusted using a spectrophotometer (530 nm) with an optical density of 0.09-0.11 to get the final concentration of 1×10<sup>3</sup> to 3×10<sup>3</sup> CFU/mL. A sterile 96-well microtiter plate with a flat bottom was used to inoculate the prepared solution. Around 100 µl of the conidial suspension in RPMI 1640 was applied to each well as an inoculant, and 100 µl of diluted medication was administered to each well in turn. Additionally, sterility and growth control wells were installed. For four days, every microtitre plate was incubated at 37°C. The control was *Trichophyton mentagrophytes* ATCC MYA-4439 [8].

### **Analytical statistics**

The mean, standard deviation, and standard error were used to measure each quantitative variable. For categorical variables, frequencies and proportions were explained. At the two-sided significance level of  $p < 0.05$ , all statistical tests that were used were evaluated. To investigate the relationship between the variables, chi-square tests were used.

### **Results**

Figure 1 shows the total number of clinical samples according to KOH and culture positivity. For skin samples, there were 111 cases, with 28 testing positive for KOH (25.22%) and 32 testing positive for culture (28.82%). Nail samples included 73 cases, with 28 testing positive for KOH (38.35%) and 32 testing positive for culture (43.83%). In total, there were 184 clinical samples, with 56 testing positive for KOH (30.43%) and 63 testing positive for culture (34.23%). The chi-square test resulted in a p-value of 0.068 for skin samples. A p-value of less than 0.05 is considered significant (Figure 1).

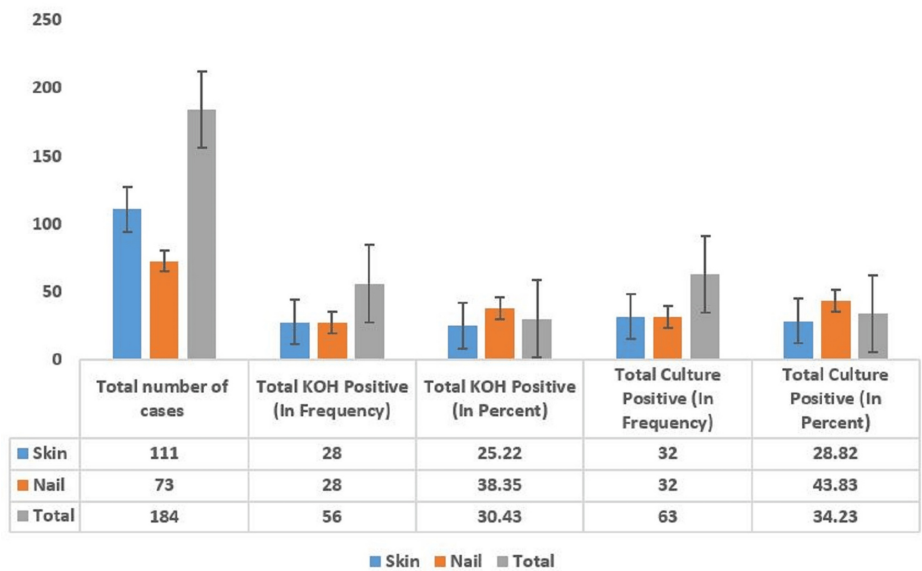


FIGURE 1: Total number of clinical samples according to KOH and culture positivity
KOH: potassium hydroxide

Table 1 presents the descriptive statistics for the minimum inhibitory concentration (MIC) of five drugs, based on 184 samples. Amphotericin showed the lowest mean MIC (0.57 µg/mL, SD 0.87), followed by terbinafine (2.71 µg/mL, SD 6.00), griseofulvin (4.13 µg/mL, SD 6.20), itraconazole (3.38 µg/mL, SD 5.89), and fluconazole, which had the highest mean MIC (7.70 µg/mL, SD 14.39). Amphotericin and terbinafine achieved 100% inhibition, while itraconazole, fluconazole, and griseofulvin showed 80% inhibition. These statistics reflect the varying effectiveness of the drugs against the tested organisms (Table 1).

| Antifungal Agents | Participants | Mean (MICs) | Std. Deviation (MICs) |
|-------------------|--------------|-------------|-----------------------|
| Amphotericin      | 184          | 0.56        | 0.86                  |
| Itraconazole      | 184          | 3.38        | 5.89                  |
| Fluconazole       | 184          | 7.69        | 14.38                 |
| Terbinafine       | 184          | 2.7         | 6                     |
| Griseofulvin      | 184          | 4.13        | 6.2                   |

TABLE 1: Descriptive statistics for selected antifungal agents MIC
100% inhibition for amphotericin and terbinafine 80% inhibition for itraconazole, fluconazole, and griseofulvin
MIC: minimum inhibitory concentration

Table 2 highlights the association of potassium hydroxide (KOH) and culture outcomes with amphotericin's minimum inhibitory concentration (MIC), analyzed using chi-square tests. KOH positivity showed a significant correlation with higher MIC values (p=0.000), as most positive cases exhibited MIC levels of 1 µL or higher. Higher MIC values were observed with specific fungal isolates, particularly T. rubrum (MIC 2-4 µL), T. mentagrophytes (MIC 1-2 µL), and T. tonsurans. This demonstrates the influence of fungal presence and species on amphotericin's MIC levels (Table 2).

| Outcome Variables |                   | Amphotericin MICs Levels |         |       |        |        |       | Chi-square | P-value |
|-------------------|-------------------|--------------------------|---------|-------|--------|--------|-------|------------|---------|
|                   |                   | 0.12 (μL)                | 0.5(μL) | 1(μL) | 2 (μL) | 4 (μL) | Total |            |         |
| KOH               | Negative          | 0                        | 2       | 1     | 5      | 0      | 128   | 11.640     | 0.000   |
|                   | Positive          | 1                        | 2       | 15    | 36     | 1      | 56    |            |         |
|                   | Contamination     | 0                        | 0       | 0     | 0      | 0      | 90    |            |         |
|                   | No fungal growth  | 0                        | 0       | 0     | 0      | 0      | 31    |            |         |
| Culture           | T. mentagrophytes | 0                        | 0       | 9     | 14     | 0      | 23    | 83.97      | 0.000   |
|                   | T. rubrum         | 0                        | 2       | 3     | 17     | 1      | 23    |            |         |
|                   | T. tonsurans      | 1                        | 2       | 3     | 3      | 0      | 9     |            |         |
|                   | T. indotineae     | 0                        | 0       | 1     | 7      | 0      | 8     |            |         |

TABLE 2: Distribution and association of KOH and culture outcomes with amphotericin MIC levels among participants

KOH: potassium hydroxide; MIC: minimum inhibitory concentration

Table 3 examines the association of KOH positivity and culture outcomes with fluconazole's minimum inhibitory concentration (MIC), revealing significant correlations (p=0.000). KOH-positive samples exhibited higher MIC values, with concentrations of 8 μL or more common. Specific fungal isolates demonstrated varied MIC distributions: T. mentagrophytes and T. indotineae were associated with higher MIC levels (16-64 μL), while T. rubrum primarily showed MIC levels of 4-8 μL. This highlights that fungal presence and species significantly influence fluconazole's MIC (Table 3).

| Outcome Variables |                   | Fluconazole MIC Levels |       |        |        |        | Total | Chi-square | P-value |
|-------------------|-------------------|------------------------|-------|--------|--------|--------|-------|------------|---------|
|                   |                   | 4(μL)                  | 8(μL) | 16(μL) | 32(μL) | 64(μL) |       |            |         |
| KOH               | Negative          | 0                      | 3     | 1      | 2      | 2      | 128   | 4.424      | 0.000   |
|                   | Positive          | 4                      | 18    | 8      | 22     | 3      | 56    |            |         |
|                   | Contamination     | 0                      | 0     | 0      | 0      | 0      | 90    |            |         |
|                   | No fungal growth  | 0                      | 0     | 0      | 0      | 0      | 31    |            |         |
| Culture           | T. mentagrophytes | 0                      | 0     | 8      | 15     | 0      | 23    | 83.53      | 0.000   |
|                   | T. rubrum         | 4                      | 19    | 0      | 0      | 0      | 23    |            |         |
|                   | T. tonsurans      | 0                      | 2     | 1      | 2      | 4      | 9     |            |         |
|                   | T. indotinae      | 0                      | 0     | 0      | 7      | 1      | 8     |            |         |

TABLE 3: Distribution and association of KOH and culture outcomes with fluconazole MIC levels among participants

KOH: potassium hydroxide; MIC: minimum inhibitory concentration

Table 4 explores the association of KOH positivity and culture outcomes with griseofulvin's minimum inhibitory concentration (MIC), showing significant results (p=0.000). KOH-positive samples had higher MIC values, with the majority falling at 8 μL or 16 μL. Specific fungal isolates displayed distinct MIC patterns: T. mentagrophytes and T. indotineae were linked to an MIC of 16 μL, T. rubrum predominantly to 8 μL, and T. tonsurans exclusively to 8 μL. This underscores that fungal presence and species significantly affect griseofulvin's MIC (Table 4).

| Outcome Variables |                   | Griseofulvin MICs Level |        |       |       | Chi-square | P-value |
|-------------------|-------------------|-------------------------|--------|-------|-------|------------|---------|
|                   |                   | 8 (μL)                  | 16(μL) | Total |       |            |         |
| KOH               | Negative          | 6                       | 2      | 128   | 2.439 | 0.000      |         |
|                   | Positive          | 25                      | 30     | 56    |       |            |         |
|                   | Contamination     | 0                       | 0      | 90    |       |            |         |
|                   | No fungal growth  | 0                       | 0      | 31    |       |            |         |
| Culture           | T. mentagrophytes | 0                       | 23     | 23    | 59.17 | 0.000      |         |
|                   | T. rubrum         | 22                      | 1      | 23    |       |            |         |
|                   | T. tonsurans      | 9                       | 0      | 9     |       |            |         |
|                   | T. indotineae     | 0                       | 8      | 8     |       |            |         |

TABLE 4: Distribution and association of KOH and culture outcomes with griseofulvin MIC levels among participants

KOH: potassium hydroxide; MIC: minimum inhibitory concentration

Table 5 highlights the association of KOH positivity and culture outcomes with itraconazole's minimum inhibitory concentration (MIC), showing significant results (p=0.000). Higher MIC levels, with concentrations of 4-16 μL being more frequent. Specific fungal isolates demonstrated unique MIC patterns: T. mentagrophytes and T. rubrum showed higher MICs, with T. rubrum mostly at 16 μL, T. tonsurans at 16 μL, and T. indotineae varying from 2-8 μL. This emphasizes the significant influence of fungal species on itraconazole's MIC (Table 5).

| Outcome Variables |                   | Itraconazole MICs Level |       |       |       |        | Chi-square | P-value |
|-------------------|-------------------|-------------------------|-------|-------|-------|--------|------------|---------|
|                   |                   | 0.5 (μL)                | 2(μL) | 4(μL) | 8(μL) | 16(μL) |            |         |
| KOH               | Negative          | 1                       | 0     | 1     | 0     | 6      | 3.913      | 0.000   |
|                   | Positive          | 7                       | 1     | 11    | 13    | 23     |            |         |
|                   | Contamination     | 0                       | 0     | 0     | 0     | 0      |            |         |
|                   | No fungal growth  | 0                       | 0     | 0     | 0     | 0      |            |         |
| Culture           | T. mentagrophytes | 8                       | 0     | 8     | 7     | 0      | 66.90      | 0.000   |
|                   | T. rubrum         | 0                       | 0     | 0     | 3     | 20     |            |         |
|                   | T. tonsurans      | 0                       | 0     | 0     | 0     | 9      |            |         |
|                   | T. indotineae     | 0                       | 1     | 4     | 3     | 0      |            |         |

TABLE 5: Distribution and association of KOH and culture outcomes with itraconazole MIC levels among participants

KOH: potassium hydroxide; MIC: minimum inhibitory concentration

Table 6 examines the association of KOH positivity and culture outcomes with terbinafine's minimum inhibitory concentration (MIC), revealing significant results (p=0.000). KOH-positive samples showed higher MIC values, with most falling at 0.06 μL or 16 μL. Specific fungal isolates displayed distinct MIC patterns: T. mentagrophytes predominantly showed an MIC of 16 μL, T. rubrum clustered at 0.06 μL, T. tonsurans ranged from 0.03-0.12 μL, and T. indotineae was consistently at 16 μL. This underscores the significant impact of fungal presence and species on terbinafine's MIC (Table 6).

| Outcome Variables |                   | Terbinafine MICs Levels |          |          |         | Chi-square | P-value |
|-------------------|-------------------|-------------------------|----------|----------|---------|------------|---------|
|                   |                   | 0.03(μL)                | 0.06(μL) | 0.12(μL) | 16 (μL) |            |         |
| KOH               | Negative          | 0                       | 6        | 0        | 2       | 4.490      | 0.000   |
|                   | Positive          | 4                       | 20       | 1        | 29      |            |         |
|                   | Contamination     | 0                       | 0        | 0        | 0       |            |         |
|                   | No fungal growth  | 0                       | 0        | 0        | 0       |            |         |
| Culture           | T. mentagrophytes | 0                       | 0        | 0        | 23      | 68.176     | 0.000   |
|                   | T. rubrum         | 3                       | 19       | 0        | 0       |            |         |
|                   | T. tonsurans      | 1                       | 7        | 1        | 0       |            |         |
|                   | T. indotineae     | 0                       | 0        | 0        | 8       |            |         |

**TABLE 6: Distribution and association of KOH and culture outcomes with griseofulvin MIC levels among participants**

KOH: potassium hydroxide; MIC: minimum inhibitory concentration

Discussion

The findings of this study highlight critical insights into the antifungal susceptibility patterns of *Trichophyton* species, as determined by minimum inhibitory concentrations (MIC) using the broth microdilution assay. These results align with and expand upon existing literature, providing both supporting and contrasting perspectives on antifungal resistance trends and species-specific MIC variations.

Amphotericin's low mean MIC (0.57 μg/mL) and 100% inhibition align with earlier reports of its potent fungicidal activity against dermatophytes [9]. However, its higher MIC values in KOH-positive samples and specific fungal isolates like *T. rubrum* and *T. mentagrophytes* reflect potential variations in resistance patterns, possibly linked to geographic differences or prior antifungal exposure [5].

For Terbinafine, its excellent efficacy and 100% inhibition support previous findings of its strong action against dermatophytes. However, its elevated MIC values in *T. mentagrophytes* and *T. indotineae* are concerning, corroborating recent reports of terbinafine resistance in these species [6]. This underscores the importance of routine susceptibility testing to monitor resistance trends.

Fluconazole's high mean MIC (7.70 μg/mL) and only 80% inhibition are consistent with its known limited efficacy against dermatophytes (Gupta et al., 2020) [10]. The significantly higher MIC values observed in *T. mentagrophytes* and *T. indotineae* mirror recent studies documenting fluconazole resistance in these isolates (Nenoff et al., 2023) [11]. These findings suggest that fluconazole may no longer be a reliable option for treating infections caused by these resistant species.

Itraconazole showed moderate efficacy with an 80% inhibition rate and a mean MIC of 3.38 μg/mL. Its higher MIC levels in KOH-positive samples and isolates like *T. rubrum* and *T. tonsurans* align with findings of species-specific variability in susceptibility [9]. This highlights the potential role of itraconazole in managing infections caused by moderately resistant strains, although its effectiveness may vary by species.

Griseofulvin demonstrated moderate activity with a mean MIC of 4.13 μg/mL and 80% inhibition. Its higher MIC values in *T. mentagrophytes* and *T. indotineae* are consistent with reports of declining susceptibility in certain dermatophyte species [5]. This trend may be linked to evolving resistance mechanisms or pharmacokinetic challenges.

The absence of gender-based differences in MIC distributions across all drugs is consistent with previous studies indicating that antifungal susceptibility is not significantly influenced by gender [4].

The significant associations of KOH positivity and culture outcomes with higher MIC values across all drugs emphasize the influence of fungal load and species diversity on antifungal efficacy. Specifically, isolates like *T. mentagrophytes*, *T. indotineae*, and *T. rubrum* consistently exhibited higher MIC values, reflecting their potential role in driving resistance trends. This aligns with recent studies advocating for species-specific susceptibility testing to optimize treatment outcomes [6,11].

## Study strengths

Clearly highlight the strengths of the study, such as the systematic use of standardized methods (CLSI) for antifungal susceptibility testing and its focus on *Trichophyton*, a significant dermatophyte genus.

## Study limitations

The study evaluated only five antifungal agents, despite the existence of numerous other antifungal agents that also exhibit resistance. The samples were collected from patients who had received prior treatment, leading to a low culture yield. Out of 184 samples, only 63 culture isolates were available for antifungal susceptibility testing, which limits generalizability due to the sample size or geographical focus.

The resistance patterns of these additional agents should be investigated in future studies with alternative methodologies, including broader sample sizes and research designs, emphasizing the importance of species-specific testing and resistance monitoring to make the findings more relevant to healthcare practitioners.

## Conclusions

In conclusion, this study sheds light on the intricate dynamics of antifungal resistance in dermatophytes, particularly concerning key antifungal agents such as fluconazole, griseofulvin, itraconazole, and terbinafine. MIC distributions play a critical role in understanding antifungal effectiveness in clinical microbiology. Results focused on unexpected resistance to typically effective drugs like terbinafine or fluconazole. As MIC-guided therapy improves outcomes by ensuring adequate drug exposure, the study results guide clinicians to choose the most appropriate antifungal for a given infection, minimizing toxicity and maximizing efficacy. These findings highlight the varying efficacy of antifungal agents against *Trichophyton* species and the critical role of species-specific and geographic resistance monitoring. Routine susceptibility testing and targeted antifungal stewardship are essential to mitigate resistance and improve therapeutic outcomes for dermatophytosis.

## Additional Information

### Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

**Concept and design:** Anshu Singh, Amber Prasad, Gagandeep Singh, Neirita Hazarika , Pratima Gupta, Neelam Kaistha

**Acquisition, analysis, or interpretation of data:** Anshu Singh, Amber Prasad, Gagandeep Singh, Neirita Hazarika , Pratima Gupta, Neelam Kaistha

**Drafting of the manuscript:** Anshu Singh, Amber Prasad, Gagandeep Singh, Neirita Hazarika , Pratima Gupta, Neelam Kaistha

**Critical review of the manuscript for important intellectual content:** Anshu Singh, Amber Prasad, Gagandeep Singh, Neirita Hazarika , Pratima Gupta, Neelam Kaistha

**Supervision:** Anshu Singh, Amber Prasad, Gagandeep Singh, Neirita Hazarika , Pratima Gupta, Neelam Kaistha

### Disclosures

**Human subjects:** Consent for treatment and open access publication was obtained or waived by all participants in this study. Institutional Ethics Committee of All India Institute of Medical Sciences, Rishikesh issued approval AIIMS/IEC/20/19, dated 08/02/2020. **Animal subjects:** All authors have confirmed that this study did not involve animal subjects or tissue. **Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

## References

1. Gnat S, Łagowski D, Nowakiewicz A, Zięba P: The host range of dermatophytes, it is at all possible? Phenotypic evaluation of the keratinolytic activity of *Trichophyton verrucosum* clinical isolates. *Mycoses*. 2019, 62:274-83. [10.1111/myc.12876](https://doi.org/10.1111/myc.12876)

2. Nenoff P, Verma SB, Vasani R, et al.: The current Indian epidemic of superficial dermatophytosis due to *Trichophyton mentagrophytes*- a molecular study. *Mycoses*. 2019, 62:336-56. [10.1111/myc.12878](#)
3. Clinical and Laboratory Standards Institute (CLSI): Reference Method for Broth Dilution Antifungal Susceptibility Testing. Clinical and Laboratory Standards Institute (CLSI), Wayne, PA; 2017. [https://clsi.org/media/1897/m27ed4\\_sample.pdf](https://clsi.org/media/1897/m27ed4_sample.pdf).
4. Lewis R, Enne VI, Ostrosky-Zeichner L: Preface. *J Antimicrob Chemother*. 2021, 76:i1. [10.1093/jac/dkaa540](#)
5. Rai M, Singh SK, Patel SN, Patel S: Regional variability in antifungal resistance among *Trichophyton* species: a systematic review. *Journal of Fungal Research*. 2022, 28:312-20.
6. Schalk E, Panse J: Which trial do we need? Scheduled central venous catheter removal vs. routine clinical care for prevention of central venous catheter-related bloodstream infections in patients with haematologic malignancies. *Clin Microbiol Infect*. 2023, 29:417-8. [10.1016/j.cmi.2023.01.003](#)
7. Negi N, Tripathi V, Chanda Choudhury R, Singh Bist J, Kumari N, Chandola I: Clinicomycological profile of superficial fungal infections caused by dermatophytes in a tertiary care centre of North India. *Int J Curr Microbiol App Sci* . 2017, 6:3220-7.
8. M38-A2 Reference Method for Broth Dilution Antifungal Susceptibility Testing of Filamentous Fungi; Approved Standard-Second Edition [Internet]. 2008 [cited 2019 Nov 26].. <http://www.clsi.org..>
9. Morelli MK, Veve MP, Lorson W, Shorman MA: *Candida* spp. infective endocarditis: characteristics and outcomes of twenty patients with a focus on injection drug use as a predisposing risk factor. *Mycoses*. 2021, 64:181-6. [10.1111/myc.13200](#)
10. Gupta A. K., Versteeg S. G., Shear N. H.: Fluconazole resistance in dermatophytes: global trends and clinical implications. *Dermatology Clinics*. 2020, 38:387-400. [10.1016/j.det.2020.03.009](#)
11. Nenoff P., Krüger C., Ginter-Hanselmayer G., Tietz H. J.: Resistance mechanisms in *Trichophyton* species: A global review of antifungal susceptibility. *Mycopathologia*, 188, 203-214. 2023. [10.1007/s11046-023-00640-7](#)