

Emerging terbinafine-resistant *Trichophyton indotineae* between 2018 and 2023: a multinational genomic epidemiology study

Johanna Rhodes*, Sui Ting Hui*, Sarah Dellièvre, Richard C Summerbell, James A Scott, Amtoj Kaur, Richard C Barton, Rodrigo Leita, Samuel Hemmings, Rebeca Goiriz, Jonathan Lambourne, Rhys A Farrer, Silke Schelenz, Roderick J Hay, Andrew M Borman, Anuradha Chowdhary, Alireza Abdolrasouli†, Matthew C Fishert



Summary

Background *Trichophyton* species cause the greatest burden of dermatophytosis worldwide, with the *Trichophyton mentagrophytes* species complex being particularly associated with the emergence of new aggressive infections. One emerging species, *Trichophyton indotineae* is notable for its clinical resistance to terbinafine antifungal treatment and rapid global spread. In this study we aim to characterise the epidemiology of this emerging pathogen using genomics.

Methods In this retrospective genomic epidemiology study, to better understand the epidemiology of this disease, we sourced isolates collected from patients with severe cases of dermatophytosis (identified either by internal transcribed spacer sequencing or phenotypic characterisation) in the UK, Ireland, France, Canada, and India for the period 2014–23, including the *T indotineae* type strain from Japan. We used whole-genome sequencing to confirm 90 isolates were *T indotineae*, and antifungal susceptibility testing to assess susceptibility to terbinafine.

Findings 103 cases of severe dermatophytosis caused by *Trichophyton* species collected between 2018 and 2023 in the UK, France, Canada, Ireland, and India were included in this study. Susceptibility testing indicated that 63 (70%) of 90 *T indotineae* isolates were resistant to terbinafine (minimum inhibitory concentration [MIC] ≥ 0.5 mg/L). Pairwise genetic distances showed very high identity with only 147 (range 1–414) single-nucleotide polymorphisms (SNPs) separating isolates that were nested within a monophyletic phylogeny, supporting a single evolutionary origin of *T indotineae*. Genome-wide analyses identified multiple non-synonymous SNPs in *SQLE* (*ERG1*), the squalene epoxidase target of terbinafine, that were associated with terbinafine in vitro resistance of 0.5 mg/L or higher. However, six isolates exhibited high MIC values without *SQLE* mutations, suggesting the presence of alternative resistance mechanisms.

Interpretation That no clear geographical clustering of isolates was observed confirms the rapid transcontinental spread of *T indotineae* from its likely centre of diversity in Asia. Our findings highlight the importance of better genomic surveillance to understand and manage this severe and rapidly emerging terbinafine-resistant dermatophyte.

Funding None.

Copyright © 2025 The Author(s). Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Introduction

Dermatophytosis is the most common fungal infection globally affecting an estimated 20–25% of the world's population.^{1,2} Infections are caused by dermatophytes, a ubiquitous and diverse group of keratinophilic filamentous fungi capable of invading human skin, hair, and nails. Dermatophyte skin infections (also known as tinea or ringworm) are characterised by annular, pruritic lesions that easily spread, and are mainly attributed to *Trichophyton rubrum* and *Trichophyton interdigitale*.³ Secondary infection from other affected body sites or fomites can occur.⁴ Most cases of superficial dermatophytosis remain localised and respond well to standard topical or oral antifungal therapy, although onychomycosis (nail infection) caused by dermatophytes is generally classed as difficult to treat.

In the past decade, *Trichophyton indotineae* (previously *Trichophyton mentagrophytes* genotype VIII), a novel and frequently terbinafine-resistant dermatophyte species, has caused an epidemic of difficult-to-treat disease in the Indian subcontinent.^{5,6} *T indotineae* is now a common infection, and often presents as widespread, inflammatory pruritic plaques on body, groin, thighs, pubic or genital area, buttocks, and the face,^{7,8} usually manifesting as chronic, recurrent, or recalcitrant disease. Skin lesions often affect several body sites and in some cases the possibility of sexual transmission was proposed.⁹ Most isolates of *T indotineae* are clinically resistant to the allylamine drug terbinafine, the widely used standard first-line therapy, and frequently exhibit elevated in-vitro minimum inhibitory concentrations (MICs; ≥ 1 mg/L) with a prevalence up to 75%

Lancet Microbe 2026;
7: 101273

Published Online January 22,
2026

<https://doi.org/10.1016/j.lanmic.2025.101273>

*Joint first authors

†Joint last authors

Medical Research Council Centre
for Global Infectious Disease
Analysis, Imperial College
London, London, UK

(J Rhodes PhD, S T Hui MRes,
R Leita MSc, S Hemmings MSc,
Prof M C Fisher PhD);

Department of Medical
Microbiology, Radboudumc,
Nijmegen, Netherlands

(J Rhodes); Department of

Tropical Disease Biology,
Liverpool School of Tropical
Medicine, Liverpool, UK

(S T Hui); Service de
Parasitologie-Mycologie,
Hôpital Saint-Louis, Assistance

Publique – Hôpitaux de Paris,
Paris, France (S Dellièvre MD);

Institut Pasteur, Immunology of
Fungal Infections, Université
Paris-Cité, Paris, France

(S Dellièvre); Sporometrics,
Toronto, ON, Canada

(R C Summerbell PhD,

J A Scott PhD); Dalla Lana School

of Public Health, University of
Toronto, Toronto, ON, Canada

(R C Summerbell, J A Scott);

Medical Mycology Unit,

Department of Microbiology,

Vallabhbhai Patel Chest

Institute, University of Delhi,

Delhi, India (A Kaur MSc,

Prof A Chowdhary MD); National

Reference Laboratory for

Antimicrobial Resistance in

Fungal Pathogens, Vallabhbhai

Patel Chest Institute, University

of Delhi, Delhi, India (A Kaur,

Prof A Chowdhary); Mycology

Reference Centre, Leeds

Teaching Hospitals National

Health Service Trust, Leeds, UK

(R C Barton PhD); Dermatology

Department, Royal London

Hospital, London, UK

(R Goiriz MD, J Lambourne PhD);
Medical Research Council Centre
for Medical Mycology,
University of Exeter, Exeter, UK

(R A Farrer PhD,
Prof A M Borman PhD);
Department of Medical
Microbiology, King's College
Hospital, London, UK
(S Schelenz PhD,
A Abdolrasouli PhD); St John's
Institute of Dermatology, King's
College London, London, UK
(Prof R J Hay DM); UK National
Mycology Reference Laboratory,
UK Health Security Agency
South-West, Bristol, UK
(Prof A M Borman)

Correspondence to:
Dr Johanna Rhodes, Medical
Research Council Centre for
Global Infectious Disease
Analysis, Imperial College
London, London W12 7TA, UK
johanna.rhodes@imperial.ac.uk

or
Prof Matthew C Fisher, Medical
Research Council Centre for
Global Infectious Disease
Analysis, Imperial College
London, London W12 7TA, UK
matthew.fisher@imperial.ac.uk

See Online for appendix

Research in context

Evidence before this study

We searched PubMed using the terms “*Trichophyton indotineae*” AND “terbinafine resistance” for articles published from database inception to Sept 2, 2024, with no language restrictions, retrieving 25 publications. These 25 publications discussed the molecular epidemiology of *Trichophyton indotineae* and reports of infections in individual countries. Five studies completed whole-genome sequencing of small numbers of isolates. None of the identified studies focused on multinational genomic epidemiology.

Added value of this study

To our knowledge, this is the first study to combine isolates from multiple countries and continents to assess the population

genomics and genomic epidemiology of the emerging dermatophyte *T indotineae*. Whole-genome single-nucleotide polymorphism analysis suggests the presence of currently under-sampled diversity in the Indian subcontinent, and an absence of geographical clustering of isolates outside the endemic area confirms rapid international transmission.

Implications of all the available evidence

Despite high burden of fungal disease and antifungal drug resistance in south Asia, this region is continuously under-represented for genomic surveillance. Given the propensity for high terbinafine resistance and morbidity, active surveillance is necessary to contain *T indotineae* and to better understand the epidemiology of this rapidly emerging dermatophytosis.

according to some studies.¹⁰ Infections thus often require subsequent prolonged treatment courses with itraconazole;¹¹ although, azole antifungal resistance can also occur.^{6,12,13} In-vitro terbinafine resistance is associated with a phenylalanine-to-leucine amino acid change at codon 397 along with other known alleles in the gene encoding the target of terbinafine, squalene epoxidase (*ERG1/SQLE*). It has been hypothesised that overuse of topical creams containing combinations of antifungals, antibacterials, and potent corticosteroids is the main driver leading to the evolution of terbinafine-resistant *T indotineae* on the Indian subcontinent.¹⁴

Since its initial description in 2020, infections caused by *T indotineae* are increasingly reported worldwide^{7,10,15,16} and spread outside of the original endemic areas has been linked to international travel.¹⁷ However, there is growing evidence of autochthonous acquisition of infection outside the previously established endemic regions.^{18,19} Given the severity and drug-resistant phenotype associated with *T indotineae* alongside a lack of understanding of the extent, ease, and mode of transmission, there is an urgent need to better understand the extent that this infection occurs outside of its endemic range. Here, we aim to report results of whole-genome sequence (WGS) analysis of a panel of *T indotineae* isolates sourced from six countries and three continents—Asia, Europe, and North America—alongside their antifungal susceptibility patterns.

Methods

Study design

We performed a retrospective genomic epidemiology study of *T indotineae* isolates collected from patients with severe dermatophytosis between 2018 and 2023 to assess the genetic basis of terbinafine resistance and population structure in relation to the *Trichophyton* genus using WGS. Any positive *Trichophyton* human isolates (identified either by internal transcribed spacer sequencing or phenotypic characterisation) from people who had severe dermatophytosis from 2018 to 2023 were collected from the UK,

France, Canada, Ireland, and India (full details of the sampling approach are in the appendix p 1). These dates were chosen due to *T indotineae* being first described in 2020; therefore, these dates cover the period immediately before description up until study conception (2023). The reference laboratories and hospitals participating in this study were St John's Mycology Laboratory, London, UK; Charing Cross Hospital, London, UK; Barts Health NHS Trust, London, UK; Mycology Reference Centre, Leeds, UK; UK Health Security Agency Mycology Reference Laboratory, Bristol, UK; King's College Hospital, London, UK; University of Alberta Microfungus Collection and Herbarium Centre for Global Microfungal Biodiversity, Toronto, ON, Canada; Saint-Louis Hospital, Paris, France; and Vallabhbhai Patel Chest Institute (VPCI), Delhi, India. Two laboratories were involved in genomic sequencing (Imperial College London, London, UK and VPCI, Delhi, India), and one laboratory for analysis (Imperial College London, London, UK).

No clinical information or baseline characteristics were collected for this retrospective, descriptive study. All clinical work, including sampling patients, isolate identification and antifungal susceptibility testing were carried out in all participating centres as part of routine standard of care, not clinical research. For whole-genome sequencing, anonymised isolates were included that were not linked to any identifiable patient information.

Clinical samples and fungal isolates

Clinicians collected specimens from patients with clinical presentation of extensive Tinea corporis or Tinea cruris. Samples were submitted to each participating laboratory as standard of care for patients. Identification and antifungal susceptibility testing of isolates were carried out as part of routine diagnostic pathway in each laboratory following local protocols. Briefly, skin scrapings were cultured on fungal-specific media including Sabouraud glucose agar supplemented with chloramphenicol and cycloheximide. Fungal cultures were incubated at 28–30°C for a minimum of 2 weeks and dermatophytes were identified by

phenotypic methods or internal transcribed spacer-based sequencing, or both. Isolates identified as *T indotineae*, *T mentagrophytes*, or *T interdigitale* were included for subsequent genomic analysis. Stocks of conidial suspensions were stored at room temperature before DNA extraction. Genomic analysis was carried out on fully anonymised isolates stored in each participating laboratory.

Antifungal susceptibility testing (AFST)

For isolates collected from the UK, Ireland, and Canada, terbinafine AFST was determined according to the CLSI M38-A2 broth microdilution method.²⁰ For Indian isolates AFST was carried out using the Clinical and Laboratory Standards Institute broth microdilution method using the M38 3rd edition guidelines.²¹ MIC for terbinafine was read at 80% inhibition of growth compared with the drug-free growth control after at least 96 h of incubation. Isolates with terbinafine MIC of 0.5 mg/L or more were considered as non-wildtype and resistant. Further details are in the appendix (p 2).

WGS and single-nucleotide polymorphism (SNP) identification

DNA libraries were sequenced using the HiSeq 4000 platform (Illumina, San Diego, CA, USA). Detailed methods of DNA extraction, purification, and preparation are in the appendix (pp 2–3). Raw Illumina reads were mapped to the *T indotineae* reference genome (GenBank GCA_023065905.1). The reference genome for the alignment of *T interdigitale* was GCA_019359935.1 and for *T mentagrophytes* was GCA_003664465.1. We used GATK HaplotypeCaller version 4.2 to call SNPs, insertions, and deletions, and used SnpEff version 5.1 to map SNPs to genes and to infer functional annotation. Annotated sequences were interrogated for non-synonymous SNPs (nsSNPs) in the *SQLE* coding area and *ERG11* gene that could be linked to terbinafine resistance. Statistical analyses to detect significant associations between mutations in *SQLE*, *ERG11B*, and in-vitro terbinafine resistance were done using the χ^2 test function in R version 4.2.3. Full methodological details are in the appendix (p 3).

Population, mating type, and phylogenetic analysis

Genetic similarity of isolates was investigated based on SNP data using principal component analysis. We calculated Weir and Cockerham fixation index (F_{ST}) values for between-country diversity. We reported the mating type for all *T indotineae* isolates based on presence of the *HMG* gene or the α -box mating type locus. We used whole-genome SNP calls to construct maximum-likelihood phylogenetic trees. An additional 65 publicly available WGS from four studies were included in this analysis to provide additional phylogenetic context.^{22–25} Further details are in the appendix (p 4).

Genome wide association study (GWAS)

We performed a GWAS using treeWAS version 1.0 to assess which loci were associated with terbinafine

resistance with a p value cutoff of 0.05 for three tests of association between terbinafine-resistant and susceptible phenotypes as a discrete vector. Further details are in the appendix (p 5).

Role of the funding source

There was no funding source for this study.

Results

A collection of clinical *Trichophyton* isolates (n=103) sourced from six countries (UK n=30, six centres; France n=23, one centre; Canada n=17, one centre; Ireland n=1, one centre; India n=31, one centre; Japan n=1, type strain; appendix p 1) and three continents recovered from patients between 2018 and 2023 were included in this study. Additionally, the collection contains the *T indotineae* type strain from Japan (figure 1A).

The median percentage of reads mapped to reference was 97.5% (IQR 95.8–98.0). The sequences from 96 (93%) of the 103 isolates were successfully mapped to the *T indotineae* reference genome (successful mapping threshold exceeding 90%), indicating reliable data for downstream analyses. Four isolates displayed low percentage mapping to the *T indotineae* reference genome (Tri-36, Tri-93, Tri-95, and Tri-97; range 3.9% to 6.2%). These four isolates also displayed low percentage mapping to the *T interdigitale* (5.7% to 6.7%) and *T mentagrophytes* (5.9% to 7.3%) reference genomes. The raw sequence data were queried using BLASTn version 2.15.0; Tri-36, Tri-93, and Tri-95 were identified as *Cordyceps fumosorosea*, and Tri-97 was identified as *Emydomyces testavorans*. Interestingly, the Tri-97 isolate also displayed raised terbinafine MIC (4 mg/L). These isolates were excluded from further analysis, due to previous misidentification (appendix p 13).

The number of high-confidence SNPs called ranged from 20 SNPs to 130 617 SNPs, with a median of 115 SNPs (IQR 59.25–139.75). Isolates with more than 100 000 SNPs called showed higher percentage mapping to either *T interdigitale* or *T mentagrophytes* reference genomes, compared with mapping against *T indotineae*. These isolates (n=8; Tri-02, Tri-04, Tri-18, Tri-39, Tri-80, Tri-84, Tri-85, and Tri-94) also displayed high bootstrap support (100%) and clustered with type genomes for *T interdigitale* or *T mentagrophytes*, and were therefore identified as either *T interdigitale* or *T mentagrophytes*. 90 isolates mapped with high fidelity to the reference *T indotineae* genome with low numbers of filtered SNP differences (range 20–346), plus the *T indotineae* type strain, confirming their correct identification as this species. Figure 1B, which includes additional publicly available *Trichophyton* WGS, illustrates the divergence of the eight non-*T indotineae* isolates along an extended branch, and further forming two different clusters within the *T mentagrophytes/T interdigitale* species complex (TMTISC). Principal component analysis on the first two principal components confirmed separation of *T interdigitale* and *T indotineae* from *T mentagrophytes* on principal component 2, with isolates from all countries

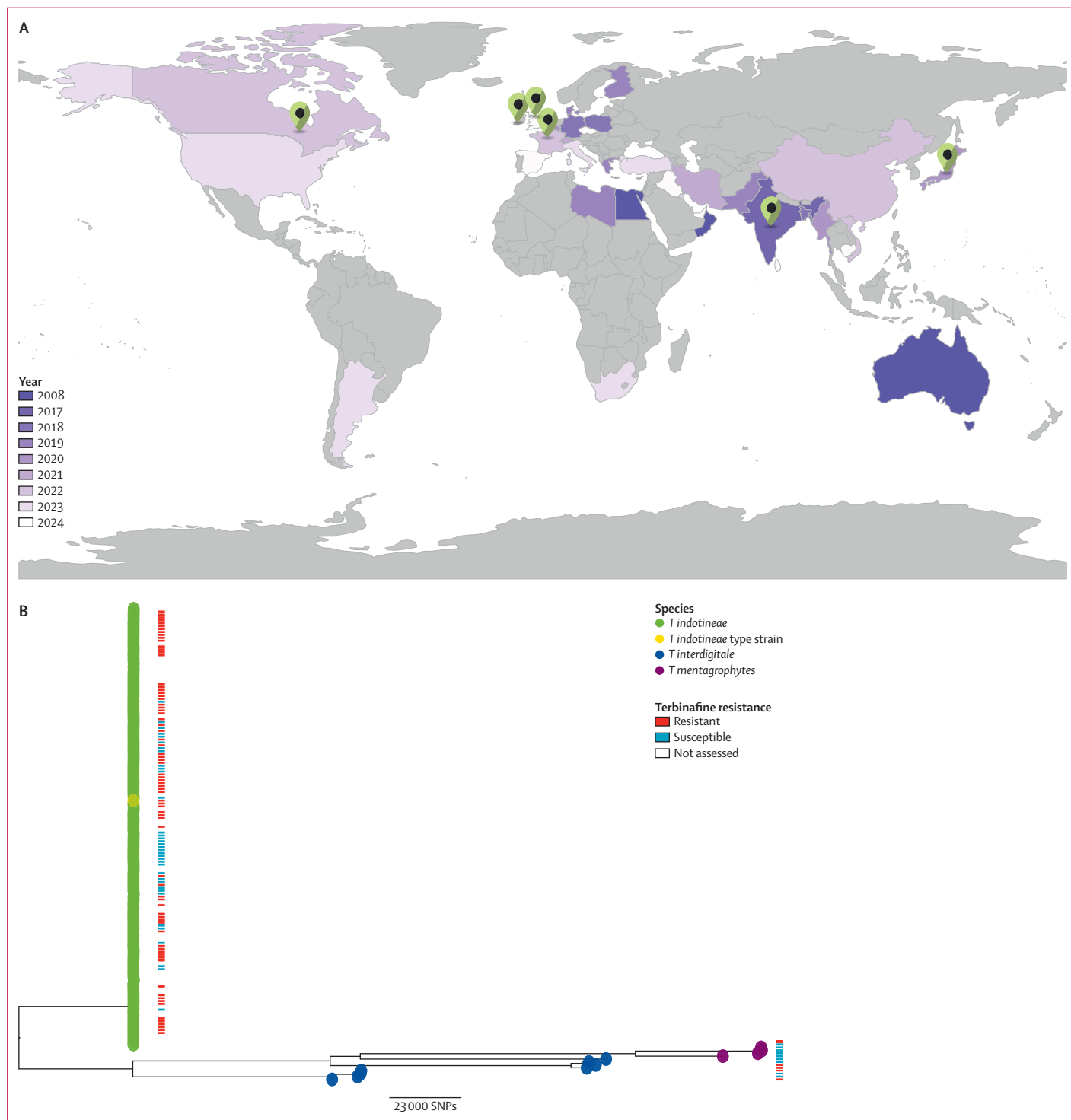


Figure 1: The global emergence of *T indotineae*

(A) Confirmed global reports of *T indotineae* indicated by year of report. Green points indicate location of sampling for isolates included in this study. (B) Genetic relationships among 102 isolates collected during this study inferred using a maximum likelihood phylogeny for isolates identified as *T mentagrophytes*, *T interdigitale*, and *T indotineae*, including the *T indotineae* type strain. Isolates with raised terbinafine minimum inhibitory concentration values, and therefore considered resistant, are indicated in red. Branch lengths represent the average number of SNPs along that branch. SNP=single-nucleotide polymorphism. *T indotineae*=*Trichophyton indotineae*. *T interdigitale*=*Trichophyton interdigitale*. *T mentagrophytes*=*Trichophyton mentagrophytes*.

occupying the same space of genetic diversity; isolates from Ireland, India, and Japan were almost indistinguishable based on principal component analysis (figure 1B).

A mean of 147 SNPs (SD 113) separates any pair of *T. indotineae* isolates (median 121 [range 59–193]). In contrast, any *T. indotineae* isolate is separated from *T. interdigitale* isolates by a mean of 231 767 SNPs (SD 12 632) and *T. mentagrophytes* isolates by a mean of 290 500 SNPs (5322), indicating substantial divergence from the TMTISC. We included the *T. indotineae* type strain CBS 146623 in this analysis, and isolates identified as *T. indotineae* were separated by a mean of 120 SNPs (95) from the *T. indotineae* type strain (figure 1B). All *T. indotineae* isolates contained the *HMG* gene, indicating the presence of MAT1-2 mating type only, confirming earlier studies.²⁶

We observed no clustering of isolates by sampling location or time (figure 2A); a principal component analysis based on whole-genome SNPs on the first two principal components for only the confirmed *T. indotineae* isolates showed isolates from all countries were drawn from the same population (figure 2B). Two lineages containing mainly terbinafine-resistant isolates sampled from the UK (n=7), Canada (n=14), the USA (n=1), and France (n=3) were observed (figure 2A); these branches were highly supported (100%; appendix p 24), indicating potentially unsampled diversity in Asia leading to the emergence of these lineages.

Within-country diversity was assessed by mean pairwise SNP distances where more than one isolate was sampled (therefore, excluding Japan and Ireland; appendix p 25). Generally, mean pairwise SNP distances were similar regardless of the country of origin (appendix p 25); we observed clusters of closely related isolates sampled from Canada, France, and the UK, whereas larger genetic diversity was observed in India (appendix p 25). Comparison of common and unique SNPs among isolates within Canada, the UK, France, and India revealed that less than 10% of SNPs were common to all four countries (appendix p 17); yet each country had a mean of 262 unique SNPs (range 200–375). Between-country diversity was assessed via the fixation index (appendix p 25); values were approaching zero in all comparisons, indicating complete sharing of genetic material. However, F_{ST} for India and Canada, and India and the UK, were higher (0.038 and 0.024, respectively), indicating some isolation between *T. indotineae* sampled in these countries.

Antifungal susceptibility testing was performed for all *T. indotineae* and non-*T. indotineae* isolates in this study. Using an MIC breakpoint of 0.5 mg/L or higher, 63 (70%) of 90 *T. indotineae* isolates were therefore determined to be resistant to terbinafine, with 22 (24%) isolates displaying an MIC of 16 or higher (appendix p 26). We therefore sought to identify the genetic basis of terbinafine resistance by performing genome-wide association using treeWAS, which accounts for confounding effects of clonality and genetic structure. treeWAS identified a single significant locus using the simultaneous test of association ($p < 0.0001$) that

mapped to the *SQLE* open reading frame, resulting in the missense mutation causing a F397L change. This statistically significant locus was present in 45 of the resistant isolates, which displayed terbinafine MIC ranges from 0.5 mg/L to 16 mg/L or more. However, we note that this mutation has been described in terbinafine susceptible *T. indotineae* isolates in Canada (figure 2A).²³ We therefore sought to further characterise the known mutations in *SQLE*, which encodes the target of terbinafine, and *ERG11*, encoding an enzyme targeted by other antifungal drug classes.

We examined the extent that polymorphisms within *SQLE* were associated with resistance to terbinafine among the 90 *T. indotineae* isolates by identifying nsSNPs conferring amino acid changes. Among the *T. indotineae* isolates, six nsSNPs were identified in *SQLE* that conferred amino acid substitutions: L393S, L393F, F397L, Y414H, S443P, and T448A. Notably, T448A (n=60; 67%) was the most prevalent substitution, whether in combination with other alleles or singular, and 80% of isolates (n=48) with this change exhibited raised MIC to terbinafine (figure 3). However, 13 (14%) isolates contained only the T448A mutation, with ten (77%) being non-resistant and three (23%) being resistant. This finding suggests that the T448A substitution is not sufficient to confer terbinafine resistance alone. The second most common substitution was F397L (n=46; 51%), and 100% of the isolates sequenced in this study with this allele demonstrated MIC values of 0.5 mg/L or higher and therefore resistance to terbinafine (figure 3). The substitution S443P (n=1; 1%), L393F (n=1; 1%), and L393S (n=4; 4%) were all found in combination with other substitutions, notably T448A, and Y414H (n=1; 1%) was found in conjunction with both T448A and F397L (figure 2A).

Although some allelic combinations were limited in occurrence, it is noteworthy that the majority of isolates with the Y414H, L393F, and L393S combination of substitutions exhibited resistance to terbinafine (figure 2A). Although the L398F substitution has been confirmed to be involved in terbinafine resistance in *Trichophyton* spp.,^{27,28} further investigation is required to understand the significance of these genotypes in contributing to terbinafine resistance. The T448A allele was of further interest because it appeared in both *T. indotineae* and non-*T. indotineae* isolates. This intriguing discovery raises further questions about the potential role of the T448A mutation in relation to terbinafine resistance.

Finally, 25 (26%) of *T. indotineae* isolates had no SNP identified in *SQLE*. 18 (72%) of these *SQLE* wildtype isolates were susceptible to terbinafine. Eight of these wildtype isolates exhibited in-vitro resistance to terbinafine with MICs 0.5 mg/L or higher, with five of these demonstrating high levels of terbinafine resistance (MIC ≥ 16 mg/L); this finding indicates the presence of alternative resistance mechanisms that do not rely on changes in *SQLE*. In the exploration of additional resistance mechanisms, an examination of the promoter region of the *SQLE* gene revealed no tandem repeats.

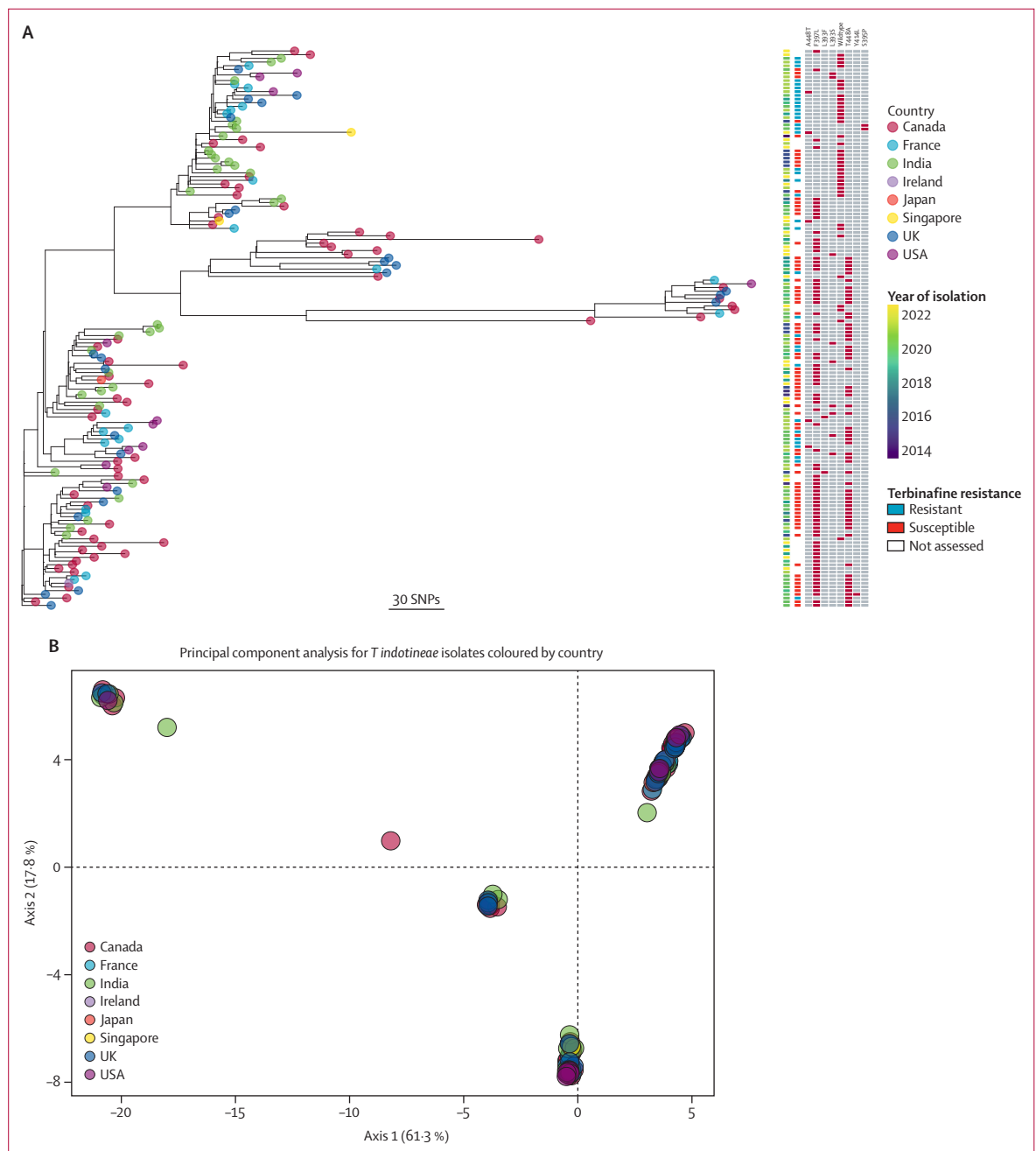


Figure 2: *T. indotineae* does not display geographical or temporal clustering

(A) Maximum-likelihood phylogeny of all confirmed *T. indotineae* isolates. Tip points indicate country of origin. Isolates with raised terbinafine minimum inhibitory concentrations, and therefore resistant, are shown in red. Year of isolation and *SQLA* amino acid substitutions are also shown. (B) Principal component analysis for all confirmed *T. indotineae* isolates shows some separation of Irish and Canadian isolates based on principal component 2. However, isolates from all countries could be seen to be drawn from the same population. SNP=single-nucleotide polymorphism. *T. indotineae*=*Trichophyton indotineae*.

Because *ERG11* is the primary target of azole antifungal drugs, we aimed to characterise mutations within this gene, and to observe any associations with mutations within *ERG11*. *T. indotineae* is thought to contain two paralogs of *ERG11*: *ERG11A* and *ERG11B*.¹² A single nsSNP conferring a R817C mutation in *ERG11A* was present in five

isolates (Tri-15, Tri-16, Tri-44, Tri-69, and Tri-73), none of which exhibited any terbinafine resistance. Of the remaining *T. indotineae* isolates, all exhibited a wild-type *ERG11A* sequence, suggesting that *ERG11A* is not involved in terbinafine resistance. Next, we analysed SNPs in the *ERG11B* sequence of *T. indotineae*, identifying S775F,

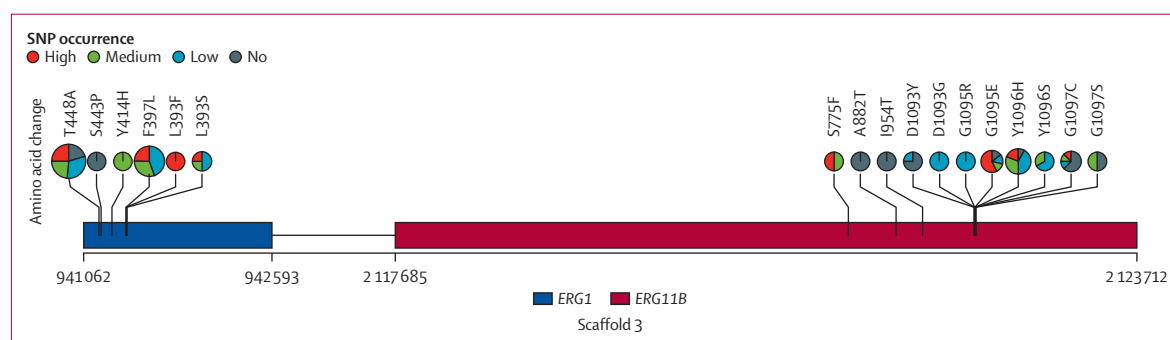


Figure 3: Amino acid location changes and corresponding SNP changes in *SQLE/ERG1* and *ERG11B* genes, present in the third scaffold (JAJVHL010000003.1) of the ASM2306590v1 assembly

Pie chart size represents the frequency of the SNP occurrence. SNP=single-nucleotide polymorphism.

A882T, I954T, D1093Y, D1093G, G1095R, G1095E, Y1096H, Y1096S, G1097C, and G1097S (figure 3). Notably, 22% (n=13) of isolates with the T448A mutation in *ERG1* also possessed the G1095E mutation in *ERG11B*. Similarly, 43% (n=26) with T448A in *ERG1* also contained the Y1096H mutation in *ERG11B*. These overlapping mutations suggest potential interactions between *ERG1* and *ERG11B*, possibly contributing to the development of terbinafine resistance. Indeed, statistical analyses (χ^2 test) highlighted significant associations between mutations in *SQLE* (p=0.0047; odds ratio 7.1 [95% CI 0.3–16.6]) and *ERG11B* (p=0.0001; 5.3 [0.2–12.6]) and in-vitro terbinafine resistance; however, we did not systematically assess these isolates for sensitivity to any azole antifungal drug. This observation underscores the need for further investigation to fully elucidate the complex genetic factors driving terbinafine resistance in individual isolates in *T. indotineae*.

Discussion

T. indotineae has become a substantial public health concern in a number of countries in the past 5 years by causing difficult-to-treat disease and displaying high levels of terbinafine antifungal drug resistance.⁶ Here, we report isolates from clinical *Trichophyton* infections from Canada, Ireland, the UK, France, and India. The WGS of 103 clinical *Trichophyton* spp isolates, including the *T. indotineae* type strain from Japan, provided important insights into the genetic basis of terbinafine drug resistance and the population structure of *T. indotineae*. Our findings highlight both the genetic diversity and the underlying mechanisms potentially contributing to terbinafine resistance, underscoring the complex nature of antifungal drug resistance in this emerging fungal pathogen.

Our WGS data reveal a high degree of genetic fidelity among 90 *T. indotineae* isolates, as demonstrated by the high percentage of reads mapped to the reference genome and low numbers of SNPs. The divergence observed in eight non-*T. indotineae* isolates emphasises the importance of accurate species identification in understanding the epidemiology and resistance patterns of fungal pathogens.

The genetic diversity of *T. indotineae* was characterised by a mean of 147 SNPs separating any pair of isolates. The addition of publicly available WGS from Singapore and the USA, and further WGS from Canada and India confirmed no clustering by geographical location or time. This lack of clustering suggests a widespread and genetically homogeneous distribution of *T. indotineae* across different regions, consistent with previous findings that indicate a high level of genetic similarity among isolates from disparate locations.¹⁸ However, assessment of within-country and between-country diversity using pairwise SNPs and F_{ST} revealed clusters of closely related isolates within Canada, France, and the UK but not India. This finding suggests that this study undersampled one likely source of *T. indotineae* (India) and has therefore not observed the true extent of genetic diversity. Future efforts to undertake large-scale global genomic surveillance would resolve this issue.

Antifungal susceptibility testing revealed that 70% of the *T. indotineae* isolates in this study exhibited in-vitro resistance to terbinafine, with 17% showing high MIC values (>16 mg/L). The genome-wide association analysis identified a single significant locus associated with terbinafine resistance, yet this locus could not fully explain the resistance observed, pointing to the involvement of additional genetic factors. Multiple nsSNPs were identified in *SQLE*, particularly the prevalent T448A mutation, and the previously reported F397L mutations.^{18,29–31} However, the presence of either of these mutations alone did not uniformly confer resistance,^{18,32} suggesting other mutations or genetic factors might modulate its impact. We identified Y414H in a single isolate with a raised terbinafine MIC (4 mg/L), which to our knowledge has not been previously reported in *Trichophyton* spp. However, a study published in 2023 reports a strain with F415C, which is close to this locus, and this polymorphism is associated with raised terbinafine MICs.³³ In addition, homology modelling shows that this mutation in the same domain of *Saccharomyces cerevisiae* confers resistance to terbinafine. Interestingly, a subset of *T. indotineae* isolates exhibited high levels of terbinafine resistance despite the absence of SNPs

in *SQLE*, indicating the existence of alternative resistance mechanisms. A study published in 2023 also identified terbinafine-resistant *T. indotineae* isolates with no known *SQLE* mutations,³⁴ and Ishii and colleagues³⁵ have suggested the TrPtk2–TrPmal pathway might play a role in promoting intrinsic terbinafine tolerance in *T. rubrum*.

Although our findings are consistent with other studies that also found *T. indotineae* isolates were wild type for *ERG11A*, we did not find previously identified *ERG11B* mutations.¹² Our analysis also suggests an association between mutations in *SQLE* and *ERG11B* and terbinafine resistance, pointing to a potential synergistic interaction between these genes in conferring resistance. Itraconazole resistance, the mechanism of which centres on mutations within *ERG11*, is rare in *T. indotineae*. Although we did not systematically carry out antifungal susceptibility to other antifungal agents in this study, future studies should consider investigating this interaction. We further acknowledge that another limitation of this study is not recording the clinical outcomes, ethnicity of the patients, or travel history, which would inform deeper epidemiological conclusions to be drawn.

Highly supported lineages identified by sampling isolates within the UK, Canada, and France suggest undersampling of the full extent of genetic diversity in Asia, despite most isolates in this study originating in India. Currently, undersampled areas include Bangladesh and Pakistan and there is limited access to WGS for countries with high prevalence. Ultimately, our findings highlight the necessity for ongoing genomic surveillance and detailed genetic analyses to monitor the spread and evolution of antifungal drug resistance in *T. indotineae*. Future research should focus on elucidating the full spectrum of genetic changes contributing to terbinafine resistance, including the potential roles of regulatory elements and non-coding regions. Linking this work with a newly established international database for drug-resistant tinea, established by the International League of Dermatological Societies in collaboration with the American Academy of Dermatology, would help to further our understanding of this growing problem.

Contributors

All authors had access to and verified the data reported in the manuscript. AA, MCF, JR and SD conceptualised the study. AA, JR, and MCF supervised the study. AA, SD, AC, AK, RCB, RCS, JAS, RG, JL, SS, RJH, and AMB conducted data collection. JR, STH, RL, SH, and RAF conducted data analysis. STH, JR, AA, and SD wrote the original draft. All authors contributed to writing, reviewing, and editing the manuscript. All authors had full access to all data presented in the study, and final responsibility for the decision to submit for publication.

Declaration of interests

AA declares honoraria for lectures given on behalf of Gilead Sciences (October, 2024). All other authors declare no competing interests.

Data sharing

All raw genomic data generated in this publication are available under the Project Accession PRJEB75499. Scripts are available at <https://github.com/mycologenomics/Trichophyton>

Acknowledgments

The UK Medical Research Council provided infrastructural support for this research. The authors would like to thank all staff in the St John's Mycology Laboratory, London, UK, for their assistance in collection and storage of clinical isolates. RAF is supported by a Wellcome Trust Career Development Award (225303/Z/22/Z). JR is supported by a Wellcome Trust Institutional Strategic Support Fund Springboard Fellowship. MCF is supported by the Wellcome Trust (219551/Z/19/Z), the Medical Research Council (MR/020258/1) and is a fellow of the Canadian Institute for Advanced Research. AC is supported by Science and Engineering Research Board (CRG/2020/001735) Department of Science and Technology, Government of India, and is a fellow of the Canadian Institute for Advanced Research.

Editorial note: The Lancet Group takes a neutral position with respect to territorial claims in published maps and institutional affiliations.

References

- Havlickova B, Czaika VA, Friedrich M. Epidemiological trends in skin mycoses worldwide. *Mycoses* 2008; **51** (suppl 4): 2–15.
- Hay RJ, Johns NE, Williams HC, et al. The global burden of skin disease in 2010: an analysis of the prevalence and impact of skin conditions. *J Invest Dermatol* 2014; **134**: 1527–34.
- De Hoog GS, Dukik K, Monod M, et al. Toward a novel multilocus phylogenetic taxonomy for the dermatophytes. *Mycopathologia* 2017; **182**: 5–31.
- Degreiff H. Clinical forms of dermatophytosis (ringworm infection). *Mycopathologia* 2008; **166**: 257–65.
- Singh A, Masih A, Khurana A, et al. High terbinafine resistance in *Trichophyton interdigitale* isolates in Delhi, India harbouring mutations in the squalene epoxidase gene. *Mycoses* 2018; **61**: 477–84.
- Singh A, Masih A, Monroy-Nieto J, et al. A unique multidrug-resistant clonal *Trichophyton* population distinct from *Trichophyton mentagrophytes/Trichophyton interdigitale* complex causing an ongoing alarming dermatophytosis outbreak in India: genomic insights and resistance profile. *Fungal Genet Biol* 2019; **133**: 103266.
- Uhrhlaß S, Verma SB, Gräser Y, et al. *Trichophyton indotineae*—an emerging pathogen causing recalcitrant dermatophytoses in India and worldwide—a multidimensional perspective. *J Fungi (Basel)* 2022; **8**: 757.
- Verma SB, Panda S, Nenoff P, et al. The unprecedented epidemic-like scenario of dermatophytosis in India: I. Epidemiology, risk factors and clinical features. *IJ DVL* 2021; **87**: 154–75.
- Spivack S, Gold JAW, Lockhart SR, et al. Potential sexual transmission of antifungal-resistant *Trichophyton indotineae*. *Emerg Infect Dis* 2024; **30**: 807–9.
- Chowdhary A, Singh A, Kaur A, Khurana A. The emergence and worldwide spread of the species *Trichophyton indotineae* causing difficult-to-treat dermatophytosis: a new challenge in the management of dermatophytosis. *PLoS Pathog* 2022; **18**: e1010795.
- Rengasamy M, Shenoy MM, Dogra S, et al. Indian association of dermatologists, venereologists and leprologists (IADVL) task force against recalcitrant *Tinea* (ITART) consensus on the management of glabrous *Tinea* (INTACT). *Indian Dermatol Online J* 2020; **11**: 502–19.
- Burmester A, Hippler UC, Elsner P, Wiegand C. Point mutations in the squalene epoxidase *erg1* and sterol 14- α demethylase *erg11* gene of *T. indotineae* isolates indicate that the resistant mutant strains evolved independently. *Mycoses* 2022; **65**: 97–102.
- Yamada T, Yaguchi T, Maeda M, et al. Gene amplification of *CYP51B*: a new mechanism of resistance to azole compounds in *Trichophyton indotineae*. *Antimicrob Agents Chemother* 2022; **66**: e0005922.
- Bishnoi A, Vinay K, Dogra S. Emergence of recalcitrant dermatophytosis in India. *Lancet Infect Dis* 2018; **18**: 250–51.
- Dellièvre S, Joannard B, Benderdouche M, et al. Emergence of difficult-to-treat *Tinea corporis* caused by *Trichophyton mentagrophytes* complex isolates, Paris, France. *Emerg Infect Dis* 2022; **28**: 224–28.
- Posso-De Los Rios CJ, Tadros E, Summerbell RC, Scott JA. Terbinafine resistant *Trichophyton indotineae* isolated in patients with superficial dermatophyte infection in Canadian patients. *J Cutan Med Surg* 2022; **26**: 371–76.

- 17 Abdolrasouli A, Borman AM, Johnson EM, Hay RJ, Arias M. Terbinafine-resistant *Trichophyton indotineae* causing extensive dermatophytosis in a returning traveller, London, UK. *Clin Exp Dermatol* 2024; **49**: 635–37.
- 18 Caplan AS, Chaturvedi S, Zhu Y, et al. Notes from the field: first reported U.S. cases of Tinea caused by *Trichophyton indotineae* — New York City, December 2021–March 2023. May 12, 2023. https://www.cdc.gov/mmwr/volumes/72/wr/mm7219a4.htm?s_cid=mm7219a4_w (accessed Dec 12, 2025).
- 19 Abdolrasouli A, Hay RJ. Antifungal-resistant *Trichophyton indotineae*: transmission is occurring outside previously identified endemic areas – are we prepared? *Br J Dermatol* 2024; **191**: 145–46.
- 20 Rex JH, Alexander BD, Andes D, et al. Reference method for broth dilution antifungal susceptibility testing of yeasts; approved standard—third edition (M27-A3). Clinical and Laboratory Standards Institute, 2008.
- 21 Alexander BD, Procop GW, Dufrense P, et al. Reference method for broth dilution antifungal susceptibility testing of filamentous fungi, 3rd Edition. Clinical and Laboratory Standards Institute, 2017.
- 22 Teo JWP, Cheng JWS, Chew KL, Lin RTP. Whole genome characterization of *Trichophyton indotineae* isolated in Singapore. *Med Mycol* 2024; **62**: myae012.
- 23 McTaggart LR, Cronin K, Ruscica S, Patel SN, Kus JV. Emergence of terbinafine-resistant *Trichophyton indotineae* in Ontario, Canada, 2014–2023. *J Clin Microbiol* 2025; **63**: e0153524.
- 24 Caplan AS, Todd GC, Zhu Y, et al. Clinical course, antifungal susceptibility, and genomic sequencing of *Trichophyton indotineae*. *JAMA Dermatol* 2024; **160**: 701.
- 25 Thakur S, Spruijtenburg B, Abhishek, et al. Whole genome sequence analysis of terbinafine resistant and susceptible trichophyton isolates from human and animal origin. *Mycopathologia* 2025; **190**: 13.
- 26 Tang C, Kong X, Ahmed SA, et al. Taxonomy of the *Trichophyton mentagrophytes*/*T. interdigitale* species complex harboring the highly virulent, multiresistant genotype *T. indotineae*. *Mycopathologia* 2021; **186**: 315–26.
- 27 Osborne CS, Leitner I, Favre B, Ryder NS. Amino acid substitution in *Trichophyton rubrum* squalene epoxidase associated with resistance to terbinafine. *Antimicrob Agents Chemother* 2005; **49**: 2840–44.
- 28 Yamada T, Maeda M, Alshahni MM, et al. Terbinafine resistance of *Trichophyton* clinical isolates caused by specific point mutations in the squalene epoxidase gene. *Antimicrob Agents Chemother* 2017; **61**: e00115–17.
- 29 Durdu M, Kandemir H, Karakoyun AS, Ilkit M, Tang C, De Hoog S. First terbinafine-resistant *Trichophyton indotineae* isolates with Phe397Leu and/or Thr414His mutations in Turkey. *Mycopathologia* 2023; **188**: 2.
- 30 Jabet A, Brun S, Normand A-C, et al. Extensive dermatophytosis caused by terbinafine-resistant *Trichophyton indotineae*, France. *Emerg Infect Dis* 2022; **28**: 229–33.
- 31 Haghani I, Babaie M, Hoseinnejad A, et al. High prevalence of terbinafine resistance among *Trichophyton mentagrophytes*/*T. interdigitale* species complex, a cross-sectional study from 2021 to 2022 in Northern Parts of Iran. *Mycopathologia* 2024; **189**: 52.
- 32 Ebert A, Monod M, Salamin K, et al. Alarming India-wide phenomenon of antifungal resistance in dermatophytes: a multicentre study. *Mycoses* 2020; **63**: 717–28.
- 33 Bortoluzzi P, Prigitano A, Sechi A, et al. Report of terbinafine resistant *Trichophyton* spp. in Italy: clinical presentations, molecular identification, antifungal susceptibility testing and mutations in the squalene epoxidase gene. *Mycoses* 2023; **66**: 680–87.
- 34 Cañete-Gibas CF, Mele J, Patterson HP, et al. Terbinafine-resistant dermatophytes and the presence of *Trichophyton indotineae* in North America. *J Clin Microbiol* 2023; **61**: e0056223.
- 35 Ishii M, Yamada T, Ishikawa K, Ichinose K, Monod M, Ohata S. The Ptk2-Pma1 pathway enhances tolerance to terbinafine in *Trichophyton rubrum*. *Antimicrob Agents Chemother* 2024; **68**: e0160923.