

# Notes on the Identification of Common Dermatophytes.

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Microscopic morphology of the micro and/or macroconidia is the most reliable identification character, but you need a good slide preparation and you may need to stimulate sporulation in some strains. Culture characteristics such as surface texture, topography and pigmentation are variable and are therefore the least reliable criteria for identification. Clinical information such as the site, appearance of the lesion, geographic location, travel history, animal contacts and race is also important, especially in identifying rare non-sporulation species like *M. audouinii*, *T. concentricum* and *T. schoenleinii* etc. Note: mating experiments are not practical for the clinical mycology laboratory.

## Identification of Common Dermatophytes

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| 1. Smooth thin-walled Macroconidia only present, no microconidia, colonies a green-brown to khaki colour.           | <i>Epidermophyton floccosum</i>                                     |
| 2. Macroconidia with rough walls present, microconidia may also be present.   | <i>Microsporum</i> Group  |
| 3. Microconidia present, smooth-walled macroconidia may or may not be present.                                      | <i>Trichophyton</i> Group   |
| 4. No conidia present, colonies sterile<br>Note: chlamydoconidia are non-diagnostic. Can you stimulate sporulation? | Non-sporulating <i>Microsporum</i> -<br><i>Trichophyton</i> Species |

*Epidermophyton floccosum* is an anthropophilic dermatophyte with a world-wide distribution which often causes tinea pedis, tinea cruris and tinea corporis. Colonies are usually slow growing, greenish-brown or khaki coloured with a suede-like surface, raised and folded in the centre, with a flat periphery and submerged fringe of growth. Older cultures may develop white pleomorphic tufts of mycelium. A deep yellowish-brown reverse pigment is usually present. Microscopic morphology shows characteristic smooth, thin-walled macroconidia, which are often produced in clusters growing directly from the hyphae. Numerous chlamydoconidia are formed in older cultures. No microconidia are formed. Key features include characteristic greenish-brown or khaki coloured cultures, the production of smooth, thin-walled, club-shaped macroconidia and the absence of microconidia.

### *Microsporum* Group

It is essential to observe macroconidia to make the identification. Difficulties occur with non-sporulating strains of *M. canis* and with the differential between *M. canis* and *M. audouinii* [use polished rice grains and potato dextrose agar]. Common species include *M. canis*, *M. gypseum*, *M. nanum* and *M. cookei*.

***Microsporum canis*** is a zoophilic dermatophyte of world-wide distribution which is a frequent cause of ringworm in humans, especially children. Invades hair, skin and rarely nails. Cats and dogs are the main sources of infection. Invaded hairs show an ectothrix infection and fluoresce a bright greenish-yellow under Wood's ultra-violet light. Colonies are flat, spreading, white to cream-coloured, with a dense cottony surface and usually have a bright golden yellow to brownish yellow reverse pigment, but non-pigmented strains may also occur. Macroconidia are typically spindle-shaped with 5-15 cells, verrucose, thick-walled and often have a terminal knob. Key features include distinctive macroconidia and culture characteristics. Abundant growth and sporulation on polished rice grains and *in vitro* perforation of hair.

***Microsporum gypseum*** is a geophilic fungus with a world-wide distribution which may cause infections in animals and humans, particularly children and rural workers during warm humid weather. Usually produces a single inflammatory skin or scalp lesion. Invaded hairs show an ectothrix infection but do not fluoresce under Wood's ultra-violet light. Colonies are usually flat, spreading, suede-like to granular, with a deep cream to tawny-buff to pale cinnamon coloured surface and a yellow-brown reverse pigment. Macroconidia are ellipsoidal, thin-walled, verrucose and 4-6 celled. Key features include distinctive macroconidia and culture characteristics.

***Microsporum nanum*** is zoophilic fungus frequently causing chronic non-inflammatory lesions in pigs and a rare cause of tinea in humans. Also present in soil of pig-yards. Infections in man usually contacted directly from pig or fomites. Invaded hairs may show a sparse ectothrix or endothrix infection but do not fluoresce under Wood's ultra-violet light. The geographical distribution is world-wide. Colonies are flat, cream to buff in colour with a suede-like to powdery surface texture with a dark reddish-brown reverse. Macroconidia are small, ovoid to pyriform, mostly 2-celled with relatively thin, finely echinulate (rough) walls, and broad truncate bases. Key features include distinctive macroconidia and culture characteristics.

***Microsporum cookei*** is a geophilic fungus which has been isolated from hair of small mammals showing no clinical lesions. Infection has been reported in rodents, dogs and rarely in humans. It is not known to invade hair *in vivo*, but produces hair perforations *in vitro*. *M. cookei* has a world-wide distribution. Colonies are flat, spreading, buff to pale brown, powdery to suede-like, with slightly raised and folded centre and some radial grooves. Reverse pigment dark reddish brown. Numerous large, very thick-walled, echinulate (rough) elliptical macroconidia with predominantly 5-6 septa but may be from 2-8 septa. Occasional spirals may be seen. Moderate numbers of mainly slender clavate with some pyriform macroconidia are present. The macroconidia are quite characteristic and diagnostic of *M. cookei* and further tests are not necessary. The thick walls and usually larger size of the macroconidia distinguished *M. cookei* from *M. gypseum*. Key features include distinctive macroconidial morphology and culture characteristics.

### ***Trichophyton* Group**

Macroconidia are less distinctive and are often absent. Microconidia are more important and their shape, size and arrangement should be noted. Culture characteristics are also useful. Common species include *T. rubrum*, *T. mentagrophytes* and varieties, *T. tonsurans* and *T. equinum*. *T. verrucosum* may occasionally produce conidia on some media.

***Trichophyton rubrum*** is an anthropophilic fungus which has become the most widely distributed dermatophyte of man. It frequently causes chronic infections of skin, nails and rarely scalp. Granulomatous lesions may sometimes occur. Infected hairs do not fluoresce under Wood's ultraviolet light, and microscopically may show endothrix or ectothrix type of invasion. Colonies are flat to slightly raised, white to cream, suede-like to downy, with a yellow-brown to wine-red reverse. Most cultures show scanty to moderate numbers of slender clavate to pyriform microconidia. Macroconidia are usually absent, however closterospore-like projections may be present in some mounts. Note: on primary isolation some cultures may lack reverse pigmentation and fail to produce microconidia. These will need to be subcultured onto media like Lactrimel agar, potato dextrose agar or *Trichophyton* Agar No.1 which stimulate pigmentation and sporulation. Key features include culture characteristics, microscopic morphology and failure to perforate hair *in vitro*.

***Trichophyton mentagrophytes* var. *interdigitale*** is an anthropophilic fungus which is a common cause of tinea pedis, particularly the vesicular type, tinea corporis, and sometimes superficial nail plate invasion. It is not known to invade hair *in vivo* but produces hair perforations *in vitro*. Colonies are usually flat, white to cream in colour with a powdery to suede-like surface and yellowish and pinkish brown reverse pigment, often becoming a darker red-brown with age. Numerous subspherical to pyriform microconidia, occasional spiral hyphae and spherical chlamydospores are present, the latter being more abundant in older cultures. Occasional slender, clavate, smooth-walled, multiseptate macroconidia are also present in some cultures. Distribution is world-wide. Key features include culture characteristics, microscopic morphology and *in vitro* perforation of human hair.

*T. mentagrophytes* var. *interdigitale* can be distinguished from *T. rubrum* and from other varieties of *T. mentagrophytes* by (a) its culture characteristics and microscopic morphology on Sabouraud's dextrose agar and/or Lactrimel agar; (b) its growth and colony morphology on Sabouraud's salt agar (colonies of *T. mentagrophytes* unlike *T. rubrum*, grow very well on this medium and usually produce a distinctive dark reddish-brown reverse pigment); (c) Other confirmatory tests useful for distinguishing *T. mentagrophytes* from *T. rubrum* include a positive urease test (within 7 days), a positive hair perforation test and the production of a yellow-brown to pinkish-brown reverse pigment on pigment stimulation media like Lactrimel and *Trichophyton* No.1 agars.

***T. mentagrophytes* var. *mentagrophytes*** is the zoophilic form of *T. mentagrophytes* with a world-wide distribution and a wide range of animal hosts including mice, guinea-pigs, kangaroos, cats, horses, sheep and rabbits. Produces inflammatory skin or scalp lesions in humans, particularly in rural workers. Kerion of the scalp and beard may occur. Invaded hairs show an ectothrix infection but do not fluoresce under Wood's ultra-violet light. Colonies are generally flat, white to cream in colour, with a powdery to granular surface. Some cultures show central folding or develop raised central tufts or pleomorphic suede-like to downy areas. Reverse pigmentation is usually a yellow-brown to reddish-brown colour. Numerous single-celled microconidia are formed, often in dense clusters. Microconidia are hyaline, smooth-walled, and are predominantly spherical to subspherical in shape, however occasional clavate to pyriform forms may occur. Varying numbers of spherical chlamydoconidia, spiral hyphae and smooth, thin-walled, clavate shaped, multicelled macroconidia may also be present. Key features include culture characteristics, microscopic morphology and clinical disease with known animal contacts.

***Trichophyton tonsurans*** is an anthropophilic fungus with a world wide distribution which causes inflammatory or chronic non-inflammatory finely scaling lesions of skin, nails and scalp. It is a common cause of tinea capitis in the Australian Aborigine and American Negro. Invaded hairs show an endothrix infection and do not fluoresce under Wood's ultra-violet light. Colonies show considerable variation in texture and colour. They may be suede-like to powdery, flat with a raised centre or folded, often with radial grooves. The colour may vary from pale-buff to yellow, the so called sulfureum form which resembles *Epidermophyton floccosum*, to dark-brown. The reverse colour varies from yellow-brown to reddish-brown to deep mahogany. Hyphae are relatively broad, irregular, much branched with numerous septa. Numerous characteristic microconidia varying in size and shape from long clavate to broad pyriform, are borne at right angles to the hyphae, which often remain unstained by lactophenol cotton blue. Very occasional smooth, thin-walled, irregular, clavate macroconidia may be present on some cultures. Numerous swollen giant forms of microconidia and chlamydoconidia are produced in older cultures. Key features include microscopic morphology, culture characteristics, endothrix invasion of hairs and partial thiamine requirement.

***Trichophyton equinum*** is a zoophilic fungus causing ringworm in horses and rare infections in humans. It is of world-wide distribution except for var. *autotrophicum* which is restricted to Australia and New Zealand. Most strains require nicotinic acid for growth except those from Australia and New Zealand, which are autotrophic (var. *autotrophicum*). Invaded hairs show an ectothrix infection but do not fluoresce under Wood's ultra-violet light. Colonies are usually flat, but some may develop gentle folds or radial grooves, white to buff in colour, suede-like to downy in texture, and are similar to *T. mentagrophytes*. Cultures usually have a deep-yellow submerged fringe and reverse which later becomes dark red in the centre. Microscopically, abundant microconidia which may be clavate to pyriform and sessile or spherical and stalked are formed laterally along the hyphae. Macroconidia are only rarely produced, but when present are clavate, smooth, thin-walled and of variable size. Occasional nodular organs may be present and the microconidia often undergo a transformation to produce abundant chlamydoconidia in old cultures. Key features include microscopic morphology, culture characteristics, nicotinic acid requirement and clinical lesions in horses.

***Trichophyton terrestre*** is a geophilic fungus of world-wide distribution which may occur as a saprophytic contaminant on humans and animals. It is not known to invade hair *in vivo*, but produces hair perforations *in vitro*. Colonies are usually flat to downy with a suede-like to granular texture resembling *T. mentagrophytes*. The surface colour may range from white to cream, buff to yellow, or greenish-yellow. Reverse pigmentation is usually yellowish-brown although some variants have a deep rose red reverse. Microconidia are large, clavate or pedicellate, usually exhibiting transition forms to more or less abundant lateral macroconidia. Macroconidia are clavate to cylindrical with rounded ends, smooth and thin-walled, and are 2-6 celled. Chlamydoconidia, hyphal spirals, racquet mycelium and antler hyphae may also be present. Key features include culture characteristics and microscopic morphology.

### **Non-Sporulating *Microsporium*/*Trichophyton* Species**

No conidia are present. Chlamydoconidia or other hyphal structures may be present but are non-diagnostic. Can you stimulate sporulation? eg for *M. canis* and *T. rubrum*. Common species include *M. audouinii*, *T. verrucosum* and *T. violaceum*. Less common species include *T. concentricum*, *T. schoenleinii*, *T. soudanense* and *M. ferrugineum*.

***Microsporum audouinii*** is an anthropophilic fungus causing non-inflammatory infections of scalp and skin especially in children. Once the cause of epidemics of tinea capitis in Europe and North America, it is now becoming less frequent. Invaded hairs show an ectothrix infection and usually fluoresce a bright greenish-yellow under Wood's ultra-violet light. Colonies are flat, spreading, greyish-white to light tan-white in colour, and have a dense suede-like to downy surface, suggestive of mouse fur in texture. Key features include the absence of conidia, poor or absence of growth on polished rice grains, inability to perforate hair *in vitro*, and culture characteristics.

***Trichophyton verrucosum*** is a zoophilic fungus causing ringworm in cattle. Infections in humans result from direct contact with cattle or infected fomites and are usually highly inflammatory involving the scalp, beard or exposed areas of the body (ie. nails, skin). Invaded hairs show an ectothrix infection and fluorescence under Wood' ultra-violet light has been noted in cattle but not in humans. Geographic distribution is world-wide. Colonies are slow growing, small, button-or-disk-shaped, white to cream coloured, with a suede-like to velvety surface, a raised centre, and flat periphery with some submerged growth. Reverse pigment may vary from non-pigmented to yellow. Broad, irregular hyphae with many terminal and intercalary chlamydospores. Chlamydospores are often in chains. The tips of some hyphae are broad and club-shaped, and occasionally divided, giving the so-called "antler" effect. When grown on thiamine-enriched media, occasional strains produce clavate to pyriform microconidia borne singly along the hyphae. Macroconidia are only rarely produced, but when present have a characteristic tail or string bean shape. All strains produce typical chains of chlamydoconidia, often referred to as "chains of pearls", when grown in brain heart infusion broth containing para-aminobenzoic acid (P.A.B.) and agar at 37°C. Microscopic examination of young 4 to 5 day old colonies, grown from a very small inoculum, on Sabouraud's' dextrose agar containing 0.5% yeast extract and incubated at 30°C, show characteristic terminal vesicles (not chlamydoconidia) at the tips of hyphae. The number of vesicles produced is greater from primary inoculations of skin scrapings or hairs. Key features include culture characteristics and requirements for thiamine and inositol, large ectothrix invasion of hair, clinical lesions and history.

***Trichophyton violaceum*** is an anthropophilic fungus causing inflammatory or chronic non-inflammatory finely scaling lesions of skin, nails, beard and scalp, producing the so-called "black dot" tinea capitis. Distribution is world-wide, particularly in the Near East, Eastern Europe, USSR and North Africa. Invaded hairs show an endothrix infection and do not fluoresce under Wood's ultra-violet light. Colonies are very slow growing, glabrous or waxy, heaped and folded and a deep violet in colour. Cultures often become pleomorphic, forming white sectors and occasional non-pigmented strains may occur. Hyphae are relatively broad, tortuous, much branched and distorted. Young hyphae usually stain well in lactophenol cotton blue, whereas older hyphae stain poorly and show small central fat globules and granules. No conidia are usually seen, although occasional pyriform microconidia have been observed on enriched media. Numerous chlamydoconidia are usually present, especially in older cultures. *T. violaceum* has a partial nutrient requirement for thiamine. There is minimal growth on casein vitamin-free agar (Trichophyton Agar No. 1), and slightly better growth on vitamin-free agar plus thiamine (Trichophyton Agar No. 4). The partial requirement for thiamine separates this organism from *T. gourvillii*, *T. rubrum*, and other species that may produce purple pigmented colonies. Key features include culture characteristics, partial thiamine requirement and endothrix hair invasion.

***Trichophyton schoenleinii*** is an anthropophilic fungus causing favus in humans. Favus is a chronic, scarring form of tinea capitis characterized by saucer-shaped crusted lesions or scutula and permanent hair loss. Invaded hairs remain intact and fluoresce a pale greenish yellow under Wood's ultra-violet light. Favus is common in Eurasia and Africa. On Sabouraud's dextrose agar, colonies are slow growing, waxy or suede-like with a deeply folded honey-comb-like thallus and some sub-surface growth. The thallus is cream coloured to yellow to orange brown. Cultures are difficult to maintain in their typical convoluted form, and rapidly become flat and downy. No reverse pigmentation is present. No macroconidia and microconidia are seen in routine cultures, however numerous chlamydoconidia may be present in older cultures. However, characteristic antler "nail head" hyphae also known as "favic chandeliers" may be observed. A few distorted clavate microconidia may be formed by some isolates when grown on polished rice grains. Key features include clinical history, culture characteristics and microscopic morphology showing favic chandeliers.

***Trichophyton soudanense*** is an anthropophilic fungus which is a frequent cause of tinea capitis in Africa. Invaded hairs show an endothrix infection but do not fluoresce under Wood's ultra-violet light. Distribution is mainly in Africa with occasional isolates from Europe, Brazil and USA. Colonies are slow-growing with a flat to folded, suede-like surface. Often there is a broad fringe of submerged growth. Surface mycelium and reverse pigment are characteristically a deep apricot-orange in colour. Microscopically, the hyphae often show reflexive or right-angle branching. Pyriform microconidia may occasionally be present and numerous chlamydoconidia are often found in older cultures. Key features include clinical history, culture characteristics and microscopic morphology showing reflexive hyphal branching and endothrix invasion of hair.

***Trichophyton concentricum*** is an anthropophilic fungus which causes chronic widespread non-inflammatory tinea corporis known as tinea imbricata because of the concentric rings of scaling it produces. It is not known to invade hair. Infections among Europeans are rare. Distribution is restricted to the Pacific Islands of Oceania, South East Asia and Central and South America. Colonies are slow growing, raised and folded, glabrous becoming suede-like, mostly white to cream coloured, but sometimes orange-brown coloured, often deeply folded into the agar with buff to yellow-brown to brown reverse. Key features include clinical disease, geographical distribution and culture characteristics.

***Microsporum ferrugineum*** is an anthropophilic fungus causing epidemic juvenile tinea capitis in humans. The clinical features are similar to those of infections caused by *M. audouinii*. Invaded hairs show an ectothrix infection and fluoresce a greenish-yellow under Wood's ultra-violet light. Reported from Asia (including China and Japan), USSR, Eastern Europe and Africa. Key mycological features include clinical history, culture characteristics and distinctive "bamboo" hyphae.

**Differentiation of *T. rubrum* and *T. mentagrophytes* var. *interdigitale* using WCH dermatophyte identification scheme.**

Medium	<i>T. rubrum</i>	<i>T. mentagrophytes</i> var. <i>interdigitale</i>
Sabouraud's dextrose agar	Colonies flat to slightly raised, white to cream, suede-like to downy with a yellow-brown to wine-red reverse. Most cultures produce scanty to moderate numbers of slender clavate to pyriform micro-conidia.	Colonies flat, white to cream, powdery to suede-like with a yellowish and pinkish brown reverse pigment. Numerous subspherical to pyriform microconidia, occasional spiral hyphae and spherical chlamydoconidia are present. Occasional slender, clavate, smooth-walled, multiseptate macroconidia may also be present.
Littman Oxgall Agar	Raised, greyish-white, suede-like to downy colony with a faint greenish-yellow diffusible pigment.	Raised white downy colonies with no reverse pigment.
Lactritmel Agar	Flat, white, downy colonies with a deep wine-red reverse pigment. Microscopically, cultures show scanty to moderate numbers pyriform to slender clavate microconidia.	Macroscopic and microscopic features as described above for SDA medium.
SDA with 5% Salt	Very stunted, white downy colony with a pale yellow-brown reverse pigment. Very poor growth.	Heaped and folded, buff-coloured suede-like surface with a dark reddish-brown submerged fringe and brown reverse. Good growth.
1% Peptone Agar	Flat, white to cream, downy colony often with a raised centre. No reverse pigment produced.	Flat, white to cream, suede-like surface with raised white downy centre. No reverse pigment.
Hydrolysis of Urea	Negative at 7 days.	Positive 5 days.
TA1 Agar	Good growth indicating no special vitamin requirements. Cultures are flat, white to cream, suede-like to downy with a deep wine-red reverse pigment.	Good growth indicating no special nutritional requirements, flat cream powdery surface with central downy tuft. Reverse pale pinkish-brown.
Hair Perforation Test	Negative at 28 days.	Positive.