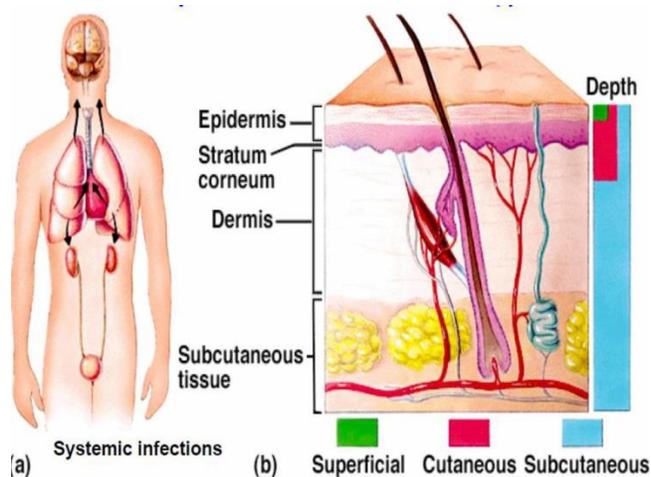


## Superficial, Cutaneous, and Subcutaneous Mycoses

Fungal infections, depending on the tissues that are initially colonized, can be classified into three major groups as follows: Superficial mycoses: These are surface infections of the skin, affecting the outermost layers of skin, hair, and mucosa. Cutaneous mycoses: These are infections of the skin involving the epidermis and its integuments, the hair, and nails. Subcutaneous mycoses: These are infections of the dermis, subcutaneous tissue, muscle, and fascia.



## Superficial Mycoses

Superficial mycosis caused by different fungi is restricted to the outermost layers of the skin and hair. The condition usually causes cosmetic problem, which can be easily diagnosed and treated. It includes four important conditions:

(a) pityriasis versicolor, (b) tinea nigra, (c) black piedra, and (d) white piedra

## Pityriasis Versicolor

Pityriasis versicolor or tinea versicolor is a superficial infection of the skin caused by *Malassezia furfur* (*Pityrosporum orbiculare*).

### Tinea Nigra

Tinea nigra is an infection of keratinized layer of skin caused by *Exophiala werneckii* or *Cladosporium werneckii*. *C. werneckii* is a dimorphic fungus that produces melanin. The fungus on the SDA grows as yeast with many cells in various stages of cell division producing typical two-celled oval structure, on primary isolation from clinical specimen. On prolonged incubation, elongated hyphae develop and in older cultures, mycelia and conidia are predominantly found.

A well-demarcated brown-black macular lesion, which appears as brownish spot of the skin, is typical manifestation of the condition. These brownish to black lesions are most commonly seen on palms and soles.

Laboratory diagnosis of tinea nigra is made by microscopy of the KOH preparation of skin scrapings collected from the affected part. Typical darkly pigmented yeast-like cells and hyper fragmented hyphae are demonstrated. Culture of the skin scraping on the SDA confirms the diagnosis.

scraping on the SDA confirms the diagnosis.

### Black Piedra

Black piedra is a superficial infection of the hair caused by *Piedraia hortae*, a dematiaceous fungus. The fungus occurs in the perfect state when it colonizes the shaft of hairs. Culture of specimens on SDA shows slow-growing brown to reddish black mycelium, which is considered asexual or anamorphic stage of the fungus. The teleomorphic state, which is the perfect state of the fungus, is occasionally found in old cultures. At this state asci, which contain spindle-shaped ascospores, develop within specialized structures. Infection of shaft of hairs of beard and scalp is the major clinical feature of black piedra. Laboratory diagnosis of the condition is made by demonstration of nodules containing asci with spindle-shaped ascospores in 10% KOH mount of the Hair.

## White Piedra

White piedra is an infection of the hair caused by yeast-like organism *Trichosporon beigelli*.

## Cutaneous Mycoses

Dermatophytoses or cutaneous mycoses are diseases of the skin, hair, and nail. These infections are caused by a homogeneous group of closely related fungi known as dermatophytes. These dermatophytes infect only superficial keratinized structures, such as skin, hair, and nail, but not deeper tissues .

## Dermatophytes

The most important dermatophytes that cause infection in humans are classified into the following three genera:

- (i) *Trichophyton*—causes infection of hair, skin, and nail.
- (ii) *Microsporum*—causes infection of hair and skin.
- (iii) *Epidermophyton*—causes infection of skin and nails, but not hair . (Fig.-1)

The dermatophytes on the basis of their natural habitat and host preferences can be classified into following groups:

- (i) **Anthropophilic species**: These dermatophytes are typically adapted to live on human host. They are transmitted from human to human through fallen hairs, desquamated epithelium, combs, hair brushes, towels, etc. Examples are *Trichophyton rubrum*, *Microsporum audouinii*, and *Epidermophyton floccosum*.
- (ii) **Zoophilic species**: These are the dermatophytes that live on animals and often cause infection in their animal host.

These zoophilic species are transmitted from infected animals to humans by direct and indirect contacts with domestic animals (e.g., cat and dog) and occasionally wild animals. Examples are *Trichophyton violaceum* and *Microsporum canis*.

(iii) **Geophilic species**: These are saprophytic fungi found in soil or in dead organic substances. They occasionally cause infection in humans and animals. Examples are *Microsporum gypseum* and *Trichophyton ajelloi*. Dermatophytes usually grow only on keratinized skin and its appendages, and do not penetrate the living tissues. In some infected persons, hypersensitivity to fungus antigen may cause secondary eruptions, such as vesicles on the finger. This reaction is known as dermatophytid (id) reaction. This reaction occurs as a result of hypersensitivity response to circulating fungal antigen, and these lesions do not contain any fungal hyphae.

Locations	Dominant Fungal species
Tinea capitis	<i>Trichophyton violaceum</i> , <i>Trichophyton tonsurans</i> , <i>Trichophyton soudanense</i> , <i>Microsporum canis</i> , and <i>Microsporum audouinii</i>
Tinea pedis	<i>Trichophyton rubrum</i> , <i>Trichophyton mentagrophytes</i> (var. <i>interdigitale</i> ), and <i>Epidermophyton floccosum</i>
Tinea cruris	<i>Trichophyton rubrum</i> and <i>Trichophyton mentagrophytes</i> (var. <i>interdigitale et granulosum</i> )
Tinea corporis	<i>Trichophyton rubrum</i> , <i>Microsporum canis</i> , and <i>Trichophyton tonsurans</i>
Tinea unguium	<i>Trichophyton rubrum</i> and <i>Trichophyton mentagrophytes</i> (var. <i>interdigitale</i> )

Fig. -1 Infections caused by different dermatophytes.

### ► Clinical syndromes

The skin infections caused by dermatophytes are chronic infections of the skin often found in the warm humid areas of the body, such as athlete's foot and jock itch. Typical ringworm lesions are circular, which have an inflamed border containing papules and vesicles surrounding a clear area of relatively normal skin. These lesions are associated with variable degrees of scaling and inflammation. Broken hair and thickened broken nails are often seen in this lesion.

Clinically, ringworm can be classified depending on the site affected. These are (a) Tinea capitis involving scalp, (b) Tinea corporis involving nonhairy skin of the

body, (c) Tinea cruris affecting groin, (d) Tinea pedis affecting foot, and (e) Tinea barbae affecting beard areas of face and neck. Favus is a chronic ringworm infection affecting hair follicle. It leads to alopecia and scarring .

### ► Laboratory diagnosis

Laboratory diagnosis is based on demonstration of fungal element in clinical specimen by microscopy and confirmation by culture. The specimens include skin scrapings and nail clippings or hair taken from the areas suspected to be infected by dermatophytes. These entire specimens are treated with alkali solution to clear epithelial cells and other debris. Direct microscopy is useful only for diagnosis, while culture is always carried out to identify the specific causative fungal agent .

### Direct microscopy

Examination of 10% direct KOH mount may show fungal hyphae. Three types of hair infections can be demonstrated in microscopy of 10% KOH wet mount as follows (Fig-2):

**Ectothrix:** Ectothrix infection is characterized by presence of a layer of arthrospores on the surface of hair shaft .It is caused by *M. audouinii*, *M. canis*, and *Trichophyton mentagrophytes*.

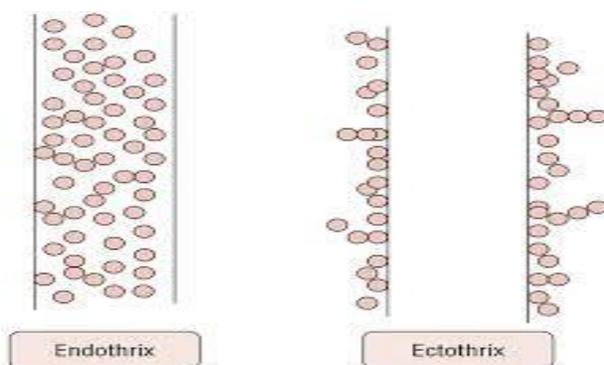


Fig.2

**Endothrix:** The clusters of arthrospores are found entirely within the hair shaft in endothrix infection. It is caused by *Trichophyton tonsurans*, *T. violaceum*, and *Trichophyton schoenleinii*.

**Favus:** In favus, there is sparse hyphal growth and formation of air spaces within hair shaft. It is caused by *T. violaceum*, *T. schoenleinii*, and *M. gypseum*

### **Culture**

The clinical specimens are cultured by inoculation on SDA containing antibiotics like cycloheximide. The media after inoculation are incubated at 25–30°C for 3 weeks. At 25°C most of the pathogenic fungi grow well, while saprophytic fungi and bacteria are inhibited.

The cultures are examined at regular intervals, and dermatophytes are identified based on (a) colony morphology, ( b) pigment production, and (c) presence of microconidia and macroconidia. The LPCB preparation of the colonies shows microconidia, macroconidia, or both. Only macroconidia are present in the *Epidermophyton* infection. Few macroconidia and more microconidia are present in *Trichophyton* infection. Macroconidia are predominantly present in *Microsporum* infection. The differentiation of three genera is based mainly on the nature of macroconidia .

### **Other tests**

**Hair perforation test:** This test is performed to differentiate *T. rubrum* from *T. mentagrophytes*. The test is also used to differentiate *M. canis* from *Microsporum equinus*. This test is performed by collecting 5–10 mm short shaft of human hair and placing it in a Petri dish with 20 mL of distilled water. Then, two

to three drops of 10% sterile yeast extract is added to the Petridish with hair shafts. These hair shafts are inoculated with test fungus cultured on SDA. The culture is incubated at 25°C for up to 1 month, during which the hairs are removed and LPCB mount of the hair is examined microscopically at weekly intervals for the demonstration of the perforation of hair.

*T. mentagrophytes* shows a positive hair perforation test characterized by a wedge-shaped perforation of the hair. This test is negative for *T. rubrum* in which only surface eruption of hair shaft is demonstrated.

**Urease test:** Urease test is carried out to differentiate *T. mentagrophytes* from *T. rubrum*. This test is performed by inoculating a tube of Christensen's medium with the fungus and incubating at 25°C for 5 days at room temperature. Most *T. mentagrophytes* are urease positive within 5 days.

**Growth on rice grains:** This test is useful to differentiate *M. canis* from *M. audouinii*. In this test, sterile, non fortified rice is inoculated with the hyphae of the fungi to be tested. The medium is observed for growth after 10 days of incubation at room temperature. *M. canis* usually grows well and forms

### Treatment

Treatment of dermatophyte infection is carried out by use of local antifungal drugs, such as miconazole, clotrimazole, econazole, etc., or by treatment orally with griseofulvin.

### Subcutaneous Mycosis

Subcutaneous mycosis is defined as fungal infection associated with development of characteristic lesion in subcutaneous tissue and overlying skin with or without extension to bone and muscle. This is caused by a heterogeneous group of fungal infection of low pathogenic potential introduced in the body percutaneously from a trivial trauma.

### Sporotrichosis

Sporotrichosis is a chronic pyogenic granulomatous lesion of the skin and subcutaneous tissue caused by *Sporothrix schenckii*. *S. schenckii* is a dimorphic fungus found all over the world. The fungus is found in soil, decaying woods, thorns, and on infected animals including rats, cats, dogs, and horse. Spore is the infective stage of the fungus. It causes infection primarily on the hand or the forearm through direct contact of the skin by spores. Typically, infection is

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introduced in skin through a penetration of thorn. At the site of thorn injury, it causes a local pustule or ulcer with the nodules along the draining lymphatics. Frequently, the regional lymph nodes draining the ulcer enlarge, suppurate, and ulcerate.

The primary lesion may remain localized or in the immunocompromised individuals may disseminate to involve the bones, joints, lung, and rarely the central nervous system. *S. schenckii* occurs in two phases. Yeast phase occurs in tissue and in culture at 37°C, while mycelium phase occurs in culture

at 22–25°C. In infected tissue, the yeast appears as round, oval, or cigarshaped cells with irregular borders. Periodic acid-Schiff (PAS) or Gomori's methenamine silver (GMS) stain is useful to demonstrate these structures in the stained smears. The fungus on SDA at 25°C produces black and shiny colonies, which become wrinkled and foggy during course of time. The mold contains hyphae bearing flower-like structures of small conidia on delicate sterigmata.

Laboratory diagnosis of sporotrichosis is made by demonstration of asteroid bodies in pus of the abscesses. Asteroid bodies consist of a central basophilic budding yeast cell with eosinophilic material, which radiates from the center.