Almost every clinically active doctor is confronted with fungal infections during their working life. It can be foot or nail fungi if they are a family doctor, fungal pneumonia if they are a haemato-oncologist, or irritant diaper dermatitis if they are a pediatrician. What matters is knowing these mycoses’ typical manifestations, diagnostic methods and therapies in order to treat patients in the best way possible. This article provides a complete insight into the wide field of mycology.

General Mycology

Characteristics of Fungi

Fungi are carbon-heterotrophic eukaryotes that form their own cell wall; they form a thallus onto or into organic material and reproduce asexually or sexually. There are more than 1,000,000 fungus species, only about 180 of which can be linked to infections in humans or animals. Besides that, fungi are used in medicine as antibiotic producers or vaccines like hepatitis B surface antigen.
The cell wall fungi form is made out of chitin, glucan and mannan. The cytoplasmic membrane of fungi possesses the sterol **ergosterol instead of cholesterol**, just like in humans. This ergosterol site is very suitable for antifungal antibiotics. As opposed to plants, fungi do not have photosynthetic capabilities and **do not possess chlorophyll**. Fungi are heterotrophs, feeding on organic materials instead.

They have more **chromosomes** and can be either haploid or diploid. Protein biosynthesis takes place on 80S ribosomes through a monocistronic mRNA. Fungi perfecti are fungi that are now in their sexual form. Those only known in their asexual form are called **fungi imperfecti** or deuteromycetes.

**Fungal Diseases**

**Allergies**

Mold antigens are the **most frequent cause of allergies**, followed by grass pollen. Some symptoms of mold allergies are headaches, losses of concentration and asthmatic complaints.

**Intoxications**

Humans can absorb mycotoxines aerogenically or through food. One of the most common toxins is **amanita phalloides** (death cap mushroom). Its **amanita toxin** leads to poisoning manifestations, like nausea and diarrhea 8–22 hours after consumption. After a few days, through a inhibition of RNA polymerase II, an enzym that helps create mRNA, cell metabolism halts and the cell will die. Another example is **claviceps purpurea**. It grows on ears of corn and this fungus’ toxin causes intestinal cramps, circulatory problems and hallucinations.

**Infections**

Infectious fungi get their pathogenicity from adherence factors, lytic **enzymes**, mycotoxins and their invasive abilities. They can be resistant to gastric acid and protect themselves from the immune system, e.g. through a slimy capsule in **cryptococcus neoformans**, or antigenic mimicry in **candida albicans**. Pathogenic fungi even develop **defense mechanisms** against specific immune cells. C. neoformans can thus induce suppressor cells, and coccidioides induces tumor necrosis factor release.

Predisposing factors for mycoses are **immunodeficiencies**. Preterm birth, malignant tumors, cytostatic therapy, corticosteroids or antibiotics, **diabetes**, kidney failure, **burns**, polytrauma, or the use of catheters and artificial respiration can all render a patient more susceptible to mycoses.

**Fungi Classification**

**Biological taxonomy according to Strassburger**

- Class I: Myxomycetes
- Class II: Chytridiomycetes
- Class III: Oomycetes
Some cells of the mycelium form fruiting bodies, the so called sporangium, in which the development of sexual conidia (or spores) takes place. If these spores are formed in a tube, the fungi are called ascomycetes. On the other hand, if they’re formed in a rack, they’re called basidomycetes. There is still no description of the sexual form of many medically relevant fungi, so they belong to fungi imperfecti.

Medical taxonomy

The classification of dermatophytes, yeast fungi and molds is more useful in medicine, although the botanic one is more precise.

<table>
<thead>
<tr>
<th>Yeasts</th>
<th>Molds</th>
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</thead>
<tbody>
<tr>
<td>Round-oval cells (budding cells = blastospores)</td>
<td>Filamentous cells (hyphen)</td>
</tr>
<tr>
<td>Pseudo micelle formation</td>
<td>Form a reticular micelle</td>
</tr>
<tr>
<td>Budding</td>
<td>Aerial micelle with fructification organs</td>
</tr>
<tr>
<td>Colony formation</td>
<td></td>
</tr>
</tbody>
</table>

Note: Identification of molds takes place microscopically through the fructification organs they formed. These reproductive structures, however, only grow in cultures, rarely in tissues.

Diagnostic Techniques

Microscopic fungus evidence
Skin scrapings, hair and nails often serve as examination materials: First, they have to be coated with KOH or sodium hydroxide to dissolve animal or human cells, while the fungal cells survive. Furthermore, many fungi are difficult to recognize under the microscope in their natural form, so they must be stained. Lactophenol or Gram staining are the simplest forms of staining.

The evidence in tissue sections is made easier to find through differential staining with periodic acid-Schiff staining (PAS reaction) or Grocott-Gomori silver staining. Optic brighteners like calcofluor, which binds with glucan and chitin on the cell wall, are also highly suitable.
Most medically relevant fungi do not have a rapid growth phase on the culture medium, but grow relatively slow. As a result, dermatophytes often need several weeks to form a visible culture. On sabouraud glucose agar or kimmig agar, fungi have a growth advantage over bacteria due to a lower pH level of the medium.

Means of differentiation are also available. Colonies of candida albicans, c. tropicalis and c. krusei thus grow on chromagar in different colours, allowing a direct identification on isolation sheets.

Biochemical yeast differentiation

Biochemical differentiation, along with the measuring of species-specific metabolic performance e.g. the utilization of different nitrogen and carbon sources or sugar
splitting, is **only performed on yeasts**. Dermatophytes and molds are distinguished through their conidia genesis.

**Serologic fungus diagnostics**

EIA, immunohaemagglutination or immunodiffusion are available as methods to detect the presence of circulating fungus-specific antibodies. However, the evidence of these antibodies is less relevant, since these antibodies can already be formed with a fungus in a mere colonization and are not always proof of an invasive infection. In immunosuppressed patients, on the other hand, antibodies are often absent, even in the case of a fungus infection.

Consequently, **positive antigen detection** does have relevance in diagnostic. Yeasts for instance release certain mannans and the capsule material of cryptococcus is composed of special glucurono-xylo-mannans. Tests on these particular fungus antigens are only positive in advanced fungus infections because macrophages only eliminate small amounts of antigens.

**Other fungus diagnostic processes**

The molecular biologic evidence of specific gene sequences in examination materials through PCR is increasingly used in diagnostic, particularly in very resistant candida phylums.

Clinical diagnosis in skin and mucosal infections is also not to be underestimated. Many dermatophytes can thus be identified through their typical manifestations on the skin.

**Specialized Mycology**

**Dermatophytes**

Cutaneous mycoses, also called dermatophytoses, are caused by dermatophytes, which are **indispensable parasites**. Dermatophytes are **keratinophilic filamentous fungi**. Infections occur through contact with infected people or as autoinfection, preferably in a warm, damp environment e.g. in the pool.

The subdivision is based on the microscopically differentiable unsexual reproductive organ forms (macro and micro conidia) in *trichophyton*, *microsporum* and *epidermophyton*.

<table>
<thead>
<tr>
<th>Fungus</th>
<th>Localization</th>
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<tbody>
<tr>
<td>Trichophyton spp.</td>
<td>Skin, hair, nails</td>
</tr>
<tr>
<td>Microsporum spp.</td>
<td>Skin, hair</td>
</tr>
<tr>
<td>Epidermophyton floccosum</td>
<td>Skin, nails</td>
</tr>
</tbody>
</table>

The dermatophytes can be transmitted via zoonotic vectors (Trichophyton spp), anthropods (Microsporum spp.), or geophillic (through the ground). All three means of transmission rely on close contact to transit the fungi.

**Note:** Immunosuppressed patients are not more prone to dermatophytes than others, but dermatophyte infections represent entrance gates for other pathogens, especially bacteria.
Symptoms of superficial dermatomycosis include dry itchy patches of skin where the fungus is present. They are called **tinea** or **ringworm**. The name of the infection is related to the body site where the dermatophyte is located: tinea capitis in the head, tinea inguinalis in the inguinal region, and tinea corporis in the stem body. In 80% of the cases, it is about a *T. rubrum* infection, in 15% about a *T. interdigitale* infection. Rarer pathogens are *M. canis* and *T. mentagrophytes*. An allergic tissue reaction is apparent among **mycoides** in dermatophytes infections.

**Tinea pedis**

*Tinea pedis*, colloquially called **foot fungus**, is the most common dermatomycosis in industrialized nations. During the course of their lives, up to 75% of the population develop symptoms of itchiness in the spaces between toes. Simultaneously, a toenail infection, called **onychomycosis**, can also often found.

**Tinea favosa**

Favus (latin for honeycomb) is a dermatomycosis caused by *trichophyton schoenleinii*. Patients have the dermatophyte in their hair. A clinical characteristic is hair infection with yellow-brownish desquamation. These scales, also called **scutula**, are pathognomonic for this disease and consist of the mycellium including hair follicles. Healing takes place through a scarring alopecia.
**Tinea capitis**

Microsporum audouinii and trichophyton tonsurans are the typical pathogens of a **head mycosis**, particularly in kids. These fungi are highly contagious and mainly infect hair. These are fully covered with spores and break off at a height of about 7 mm. Mucosa is not infected.

**Therapy for dermatomycosis**

For local therapy, topical imidazole **antimycotics** in the form of lotions or nail polish. Patients are also encouraged to keep the affected area dry. Treatment lasts about 4-6 weeks. In some cases, a systemic therapy with azolen or griseofulvin can be used as a supplement. The systemic has a longer duration than topical therapy because it takes time for antifungals to reach the keratinic layer of the skin.

**Dermatomycoses prophylaxe**

**Nail and skin care** is very important, especially in people with underlying immunosuppressive diseases like diabetes mellitus. Care should be taken for these patients to avoid warm, damp room, like saunas and check their skin for evidence of infection.

**Yeasts**

Yeasts multiply through budding. This means that a bud grows from the mother cell through evagination, with an ingrown copy of the cell’s nucleus. The so formed daughter cell eventually becomes independent.

**Candida albicans illnesses**

Candida spp. is a dimorphic fungus that can form a pseudomycelia. They are part of the saprohythic **human local flora**. Thus candida strains are detectable in test materials from the oropharynx (30 %) and the feces (65 %). Candida species are **possible pathogens**, in certain conditions.

Favoring factors of yeast colonization are:
- Reduction of physiological bacterial flora on skin and mucosa due to antibiotics
- pH level increase in the vagina or estrogen excess due to hormonal contraceptives or pregnancy
- Skin barrier damages due to burns
- Immunosuppression e.g. in AIDS, radiotherapy or cytostatic therapy

Where it’s damp, warm and dark, like in the spaces between toes or adiposities in skinfolds, inflammatory, flush lesions arise and can macerate and tear, forming rhagades. They are called **intertrigo**. Candida infections can form in these areas.

![Image: “Thrush in a child who had taken antibiotics” by James Heilman, MD. Licence: CC-SA BY 3.0](image)

A **vulvovaginal Candidosis** can arise in women, especially if they’re taking hormonal contraceptives or during pregnancy. Typical symptoms are itching and burning, along with whitish fluor vaginalis. In men, it’s called **balanitis**. In babies, haematologic patients and after antibiosis a whitish layer can be formed on the oral mucosa. Typical presentation of oral thrush is a white fluffy layer that can be wiped away.

Invasive or **systemic candida mycoses** can lead to **direct or hematogenous sepsis**. Typical consequences are **microabscesses** in liver, kidneys and lungs. However, uveitis, meningitis, arthritis and pericarditis are also a possibility.
Evidence of candida

Evidence of the candida genus can mostly already be found microscopically in the native preparation or after **gram staining**. A good representation can also succeed after **Grocott-Gomori** impregnation with silver salts or after treatment with optic brighteners like calcofluor. Typically, **cream coloured, porcelain-like colonies** develop in culture.

**N.B:** Evidence of candida in skin swabs, sputum, urine and solid faces is not proof of an infection, but only the expression of a colonization. What matters is quantity. If detection via breeding and microscopy is not successful, fungus antigen evidence can be useful; the sensitivity of these antigen tests and of anticorp tests lies at only 20-80% and its specificity is unknown.

**Candidiasis therapy**

Treatment of candidiasis is conducted through topical application fluconazole or topical imidazole. Oropharyngeal and vulvovaginal infections are both treated with local and systemic antimycotics. In systemic mycoses, the infectious cause, must first be removed, then therapy begins with high-impact triazole and echinocandin.

**Cryptococcus neoformans**
Cryptococcus neoformans is a yeast-like fungus with a **polysaccharide capsule**. The capsule protects it from phagocytose through granulocytes and macrophages. The fungus occurs in the ground, and can be found especially in **pigeon feces**. It is transmitted via respiratory droplets. Typically, the fungus will go unnoticed in the immunocompetent host. However, in immunocompromised patients, such as AIDS patients, the fungus can undergo hematological spread to the brain. 10% of all AIDS patients will develop Cryptococcus.

Patients with cryptococcal meningitis will have symptoms of nausea, headache, confusion, and cranial nerve deficits. Diagnosis occurs via a lumbar puncture, where the CSF will be stained with **India Ink**. The polysaccharide capsule will pick up the dye and a diagnosis can be made.

Treatment is a **triple combination** (amphotericin B, 5-fluorocytosine, fluconazole). The therapy must be conducted over a period of 4-8 weeks. Male patients also need a **lifelong maintenance therapy** because reactivations from the prostate can occur.

Cryptococcus can also cause pneumonia, skin ulcers, and bone lesions.

**Malassezia furfur**
Malassezia furfur is the pathogen for **pityriasis versicolor** or **tinea versicolor**, which is a superficial infection of the skin. Tinea versicolor presents itself clinically through hypopigmentation of the infected area. In rare cases, malassezia furfur can also determine a **catheter associated sepsis** in parenteral feeding with lipid solutions.

Diagnosis can be made with skin scrapings and KOH under a microscope. Treatment includes dandruff shampoo applied to the area. Topical antifungals can also be used in severe cases.

**Aspergillus**

Its typical micromorphological characteristic is conidia carriers ending in a vesicula.

This fungus can cause three different conditions: Allergic Bronchopulmonary Aspergillosis (ABPA), aspergilloma, and invasive aspergillosis. Inhalation of aspergillus spores can cause an allergic reaction and mediate the release of IgE antibodies. This is a type I hypersensitivity.

In patients with previous episodes of tuberculosis with lung cavitations, inhaling aspergillus spores cause a aspergilloma, which is a localized ball of fungus that might have to be surgically removed.

In immunocompromised patients, an aspergillus infection can disseminate and cause frequent bleeds and a bloody cough secondary to aspergillus lung nodules. This disease is very fatal and treatment is with strong antifungal medications.

Aspergillus can also create a toxin called the aflatoxin. This toxin grows on grains and peanuts. If ingested in large quantities, this toxin causes liver damage that can lead to liver cancer.
Zygomycetes

Zygomycetes are primitive dermatophytes, among which mucorales like *rhizopus* and *mucor* are particularly important in human medicine. These ubiquitous fungi are typical possible pathogens in *neutropenia* with vessel affinity. The illness can present itself as:

- **Cutane mycosis:** e.g. in extensive burns
- **Rhinocerebral mycosis:** Originates in a colonization of respiratory tract and paranasal mucosa. The spreading into the CNS can occur especially in diabetic metabolic conditions (ketoacidose).
- **Pulmonary mycosis:** Especially in leukemic patients; through ingrowth in lung vessels it can lead to lung infarct.
- **Gastrointestinal mycosis:** The fungus can grow inside the vessels and lead to intestinal infarct.
These fungi have affinity for the sinuses and cranial bones. These are highly likely to infect diabetics, burn patients, and people to take iron chelators.

**Dimorphous fungi**

Dimorphous are fungi that grow as yeasts in their parasitic form and as filamentous fungi in their saprophytic form. They are pathogens for the classic **system mycosis**.

**Histoplasma capsulatum**

This fungus lives in dry, hot climates and is confined to endemic areas in South America, USA and Africa. Its spores are spread through **bird and bat faeces-contaminated dust**. Since histoplasma capsulatum is highly contagious, it is classified in risk group III.

**Note:** It is not transferrable from human to human.

After inhalation, microconidia are phagocytized and multiply themselves in alveolar macrophages. This leads to the symptoms of histoplasmosis. This illness often progresses subclinically, but it can also turn into acute **pneumonia**. In immunosuppressed patients, there is a danger of haematogenous metastasis in **lymph nodes**, liver, **spleen** and bone marrow.
In x-rays, histoplasma granulomas appear as **nodules**. They are also called “blizzard” or “cloudburst”. Breeding takes relatively long and since histoplasma can grow again in the breeding ground as filamentous fungus, there is a high risk of infection for the lab personnel. Serologic antigenic evidence is more suitable.

**Blastomyces dermatitis**

Blastomyces dermatitis is the pathogen for North American blastomycosis. The infection takes place through inhalation or transcutaneously through skin wounds. **Tuberculosis-like symptoms** and spreading in bones and skin with formation of fistulas can occur. The developing granulomatous nodules ulcerate and heal with a central scar. Without therapy, the **lethality** of blastomycosis is **high**. Amphotericin B is indicated for treatment.
Note: Men are affected by blastomycosis 10 times more often.

Pneumocystis jiroveci

Pneumocystis is a peculiar fungus, because, unlike other fungal cells, its cells do not possess ergosterol in their cytoplasmic membrane. Because of this, it is resistant to antifungals. Pneumocystis-pneumonia is an endogen reinfection in a weakened immune system. About 2/3 of the population go through unapparent infections as long as 3 years, during which the pathogen stays in their lungs as saprophyte. In up to 80% of AIDS patients, an infection occurs.

Pneumocystes can be detected directly in cysts, plasmatic cells that were attacked in the interstitium (plasmatic cell pneumonia).

This disease is lethal if not treated. The therapy should consist of cotrimoxazole or pentamin-isethionate, and prophylaxe is also important in AIDS patients and highly immunodeficient patients.
Review Questions

The correct answers can be found below the references.

1. A 55-year old man is taken to hospital due to a severe pneumonia. He says he has been prescribed unacid by his family doctor but his symptoms have not gotten better after a week of treatment. You find out in the anamnesis that the patient spent a holiday in Texas three weeks ago. You conduct a bronchoscopy with bronchoalveolar lavage for microbiological evidences and begin an antibiosis with tazobac and piperacillin. Two weeks later, not only the patient’s symptoms have not improved, you also receive a call from the microbiologic laboratory, saying that a colleague got infected from the culture per inhalationem. Which pathogen is the most likely cause?
   A. Pneumocystis jiroveci
   B. Streptococcus pneumoniae
   C. Aspergillus fumigatus
   D. Histoplasma capsulatum
   E. Mycoplasma pneumoniae

2. What is indicated in the therapy against a pneumocystis jiroveci pneumonia?
   A. Cotrimoxazol
   B. Fluconazole
   C. Amphotericin B
   D. Caspofungin
   E. Griseofulvin

3. A 27-year old man comes to your family doctor’s practice because he noticed a whitish layer in his throat area. You can easily remove the layer during your inspection and you see spore cells in the microscopy. Which examination is indicated for this otherwise healthy man?
   A. Aerobic and anaerobic blood culture
   B. Anticorps test on candida spp.
   C. HIV test
   D. Microbiologic culture and resistance testing
   E. None of the above

References


Correct answers: 1D, 2A, 3C