Micromycetes

Molds

Microscopic fungi
Micromycetes - microscopic fungi - molds microfungi
PROKARYOTES

ARCHAEA
- Methanogens
- Extreme halophiles

BACTERIA
- Proteobacteria
- Gram-positive bacteria
- Mitochondrion
- Cyanobacteria
- Chloroplast
- Hyperthermophiles

EUKARYA
- Plants
- Slime molds
- Fungi
- Flagellates
- Animals
- Giardia
- Eukaryotic "Crown species"

Root of the tree
Alternative system
Fungi

- Kingdom of **Eukaryota**
- Eukaryotic organisms without plastids
- Nutrition absorptive (osmotrophic)
- Cell walls containing **chitin** and **β-glucans**
- **Mitochondria** with flattened cristae
- Unicellular or filamentous
- Mostly non flagellate
- Reproducing **sexually** or **asexually**
- The **diploid phase** generally short-lived
- Saprobic, mutualistic, or parasitic
Size of micromycetes

• 1.5 milions species, only 5% of them were formally classified
• Great diversity of life cycles and morphology
• Recent taxonomy is based on DNA analysis
• Majority of micromycetes is growing as fibre (hypha), that are cylindrical, fibrous structures 2–10 μm in diameter and long up to several centimeters.
Kingdom Fungi

Division:
- Chytridiomycota
  - Microsporidiomycota
  - Zygomycota
- Zygomycota
- Glomeromycota
- Ascomycota
- Basidiomycota
Fungi and pseudofungi

Kingdom: **PROTOZOA**

- *Myxomycota*

Kingdom: **CHROMISTA**

- *Plasmodiophora brassicae*
- *Spongospora subterranea*
- *Phytophthora infestans*
Fungi and pseudofungi

Kingdom: **PROTOZOA**  Division

Acrasiomycota
Myxomycota
Plasmodiophoromycota

Kingdom: **CHROMISTA**  Division

Labyrinthulomycota
Peronosporomycota
Hyphochytriomycota

Kingdom: **FUNGI**  Division

Chytridiomycota
Microsporidiomycota
Glomeromycota
Zygomycota
Ascomycota
Basidiomycota
Fungal ecology

- **saprobic**
  - Penicillium sp.

- **mutualistic symbioses**
  - Mycorrhizal fungi

- **parasitic**
  - dermatophyte
  - Cordyceps lloydii
  - Mycosphaerella fragariae
  - Spinellus fusiger
  - + endophyte
  - lichens
Kingdom: Fungi (Ophisthokonta)

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*Nosema apis*
Kingdom: Fungi (Ophisthokonta)

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*Rhodotorula* sp.
Structure of hyphae tip
Growth tip of hypha
Mikromycety - buňka

- **Vacuole**: cytoplasm less dense in older parts
- **Growing tip**
- **Nuclei**: this hypha is coenocytic (aseptate)
- **Cell wall**
- **Cell membrane**
- **Golgi apparatus**
- **Mitochondrion**
- **Rough endoplasmic reticulum**
- **Nucleus**
Cell wall – chemical composition

• Polysaccharides
  - **chitin** (N-acetylglucosamin)
  - **chitosan** (deacetylated chitin)
  - **β-glucans, mannans**
  - next polysaccharides formed from 6-deoxyhexose
    (e.g. rhamnose -6-deoxy-L-mannosa, etc.)
  - **cellulose**
  - compounds resembling **lignin** (support the wall rigidity)

• Proteins
• Fats
• **Waxes** (respond for minimal moistening)
Cytoplasmatic membrane

- Responsible for transport of nutrients and osmoregulation
- Location of some components of cell wall synthesis
- Oxidative phosphorylation does not take place in membrane
- Invaginations are not so common as in yeasts
Nucleus

- Double membrane with huge pores
- Position in the centre of the cell
- Cells of some hyphae are **coenocytic** (multinuclear – a)
- Spores contain only one nucleus
- Number of chromosomes in haploid nucleus – 7 to 40
Basic cytoplasm

- Composition and function does not differ from yeast cytoplasmy
- Main storage compound are lipids
- Lipids are stored in vacuols, in cytoplasm
- In older cells the lipids may release from vacuols, may be considered for spores
Main organels

- Nucleus
- Mitochondria (mtDNA)
- Endoplasmatic reticulum
- Golgi apparatus
- Vacuols
Mycelium

- Spore
- Hypha
- Progressive branching
- Mycelium – tangle of hyphae
- Scleromycelium – hard globular structure made from dense tangle of hyphae
- Stroma – leatherlike tangle of hyphae
Mycelium
substrate and aerial

Aerial mycelium
Substrate mycelium
agar
Two types of hyphae

• Zygomycota and Chytridiomycota – vegetative mycelium **is not septated**

• Ascomycota and Basidiomycota – vegetative mycelium is septated, may be perfored (ascomycetes). Dikaryotic basidiomycetes are formed **přesky**.
Reproduction of micromycetes

- Vegetative
- Sexual
Reproduction of micromycetes - vegetative

• Outgrowth of hyphae
• Vegetative spores are formed on
  - vegetative hyphae
  - fructification organs
    -- exospores
    -- endospores
Vegetative spores on
- vegetative hyphae

a – oidie (arthrospores) are formed by desintegration of hyphae
Shape and placement of exospores

Microconidia and macroconidia

- a – individual spores on septated mycelium (*Sporotrichum*)
- b – false bulb from chain of spores (*Cephalosporium*)
- c – halfmoon macroconidia (*Fusarium*)
- d – chain of septated macroconidia (*Alternaria*)
- e – spiral macroconidia (*Helicoma*)
- f – star shape macroconidia
Reproduction of micromycetes – vegetative – endospores in sporangium

a – without columella (Mortierella)
b – with columella (Mucor)
c – columella with apophyse (Rhizopus)
d – tubular sporangia (Piptocephalis)
e – sporangioles (Thamnidium)

1 - sporangium
2 - sporangiofor
3 - kolumela
4 - apofýza
5 – bazální buňka
Life cycle

- **ascomycota**
- **pleomorphic life cycle**
- **anamorph**
  - conidia
  - conidiophore
- **teleomorph**
  - ascus
  - ascospore
- **ascocoma**
- **teleomorph**
  - ascomata
  - Ascus
- **HOLOMORPH**
Life cycle of zygomycetes (*Rhizopus*)

1. Aerial hypha produces a sporangium
2. Sporangium bursts to release spores
3. Spore germinates to produce hyphae
4. Vegetative mycelium grows
5. Gamete forms at tip of hypha
6. Plasmogamy
7. Zygosporangium forms
8. Zygosporangiospore forms
9. Zygote produces a sporangium
10. Spores are released from sporangium
11. Spore germinates to produce hyphae
Division ASCOMYCOTA

• Subdivision: Taphrinomycotina (syn. ARCHIASCOMYCOTINA)
  Class: Schizosaccharomycetes
  Class: Taphrinomycetes
• Subdivision Saccharomycotina
  Class: Saccharomycetes
• Subdivision Pezizomycotina (syn. ASCOMYCOTINA)
  Class: Laboulbeniomyces
  Class: Eurotiomycetes
  Class: Pezizomycetes
  Class: Leotiomyces
  Class: Lecanoromycetes
  Class: Sordariomycetes
  Class: Dothideomycetes
Life cycle of **ascomycetes**

(Eupenicillium)
Life cycle of ascomycetes
Mycotoxines

- Mycotoxines are toxic secondary metabolites of many species of microscopic filamentous fungi (molds), that can contaminate a broad spectrum of food and feed. Producers of these dangerous natural contaminants cause various toxic syndromes called mycotoxicoses.
- Action of mycotoxines depends on type of toxin, duration of its effect, dose and age, its nutrition and actual state of health.
- Target organs of mycotoxines are on the first place the cells of liver, kidney, lungs, and nerves, endocrinous glands and cells of immune system. They can cause an acute toxic reaction and some can have mutagenic, terratogenic, carcinogenic and estrogenic effect.
Surveillance of mycotoxins

- Numerous international authorities are trying to reach worldwide standardization of **regulatory limits for mycotoxines**. Recently, more than 100 countries set up regulatory limits for mycotoxines in feed industry.

- Today **13 mycotoxines** or groups of mycotoxines are recorded. Evaluation process necessary for regulation of mycotoxines include a great complexes of laboratory tests, that use extraction, purification and separatory techniques.

- Majority of official control methods is based on high pressure liquid chromatography (**HPLC**).

- **Many standards for mycotoxines analysis is guaranteed by European Committee for Standardization** (CEN).
STRUCTURE of AFLATOXINES

Aflatoxin B1

Aflatoxin B2

Aflatoxin G1

Aflatoxin G2
Fusarious mycotoxines

- Fusaria belongs to noticeable originators of mycoses at human and produce mycotoxines threatening the health. Among the most dangerous mycotoxines are trichotecens, zearalenon, fumonizins.
- Zearalenon ($C_{18}H_{22}O_5$) is white, or pale yellow crystallic compound without odour. Temperature of thawing about 161 - 164 °C. Insoluble in water, tetrachlormethane ($CCl_4$) and alcohols.
- Zearalenon is relatively lipophilic compound. Its content is reduced during technology of processing of cereals.
Zearalenon

- Isolated in 1966
- White or pale yellow crystalic compound
- Temperature of thawing 161 – 164 °C
- Common in flour and cereal products.
- Estrogenous effect
Fumonisines

- Six structures: FB$_1$, FB$_2$, FB$_3$, FB$_4$, FA$_1$ a FA$_2$
- Derived from unsaturated fatty acids
- Disorders of sphingolipid metabolism
- Liver and kidney tumors at rodents
Trichothecenes

- 80 species (T-2 toxin, deoxynivalenol-DON…)
- Chemically variable
- Tricyclic seskviterpenes
- Signs of intoxication: inflammation of gastrointestinal tract, nausea, diarrhea, inhibit proteosynthesis, impaired immune system

![Structure of deoxynivalenol and T-2 toxin]
Ergot alkaloids

• An **ergot kernel**, called a **sclerotium**, develops when a **spore** of fungal species of the genus *Claviceps* infects a floret of flowering grass or cereal. The infection process mimics a **pollen grain** growing into an **ovary** during **fertilization**.

• **Ergot alkaloidy are** produced as a mixture of toxic alkaloids in **sklerotium** of some *Claviceps* species, that are common pathogens of grass.

After consumption of ergot sklerotium from infected cereals, usually as a bread baked from contaminated flour, **ergotismus** took place, human disease historically known as the **Saint Antony’s Fire**.

**Two forms of ergotism**

1) gangrenose affecting the blood circulation in limbs
2) spasmic, affecting **central nerve system**.

Modern methods of cereal cleaning reduced significantly **ergotism** as human disease, but it is still a veterinary problem.

Ergot alkaloids have pharmaceutical application.
Claviceps purpurea
Ergot alkaloids

- Ergot alkaloids are classified as:
- derivatives of 6,8-dimethylergoline and
- lysergic acid derivatives
- The neurotropic activities of the ergot alkaloids may also cause hallucinations and attendant irrational behaviour, convulsions, and even death
Aspergillus

- frequently causes contamination of food and feed
- manufacture of organic acids
- mycotoxine (Aflatoxins)
- osmophilic, osmotolerant
- allergens
- opportunistic pathogen
- warmer parts of the world
**Aspergillus niger**

Black aspergilli (*Aspergillus* section *Nigri*)

- Colonies consisting of a compact white or yellow basal felt with a dense layer of dark brown to black conidiophores

Conidial heads black, radiate. Conidiophore stipes smooth-walled

Conidia globose to subglobose

Important (toxic) metabolites: naphto-γ-pyriones, malformins, **ochratoxin A** (few isolates)

Conidia globose to subglobose
Penicillium

- Over 300 species
- One of the most widespread fungus
- Frequently causes contamination of food and feed
- mycotoxine
- allergens
- Pathogen – only one species *P. marnefei*
- Food processing (*P. camemberti, P. roqueforti, P. nalgiovense*)
- Antibiotic (Penicilin)
**Penicillium expansum**

Apple infected with *Penicillium*

Colonies on MEA after one week

Exudate absent

Odour aromatic-fruity

Conidiophores and conidia

Important (toxic) metabolites: roquefortine C, patulin
Fusarium (tel. Giberella, Nectria)

The most important mycotoxin
1. Aflatoxiny B a G
2. Aflatoxin M1
3. Patulin
4. Ochratoxin A
5. Deoxynivalenol (DON)
6. Zearalenon
7. Fumonisiny
8. T-2 toxin
Cladosporium

A very common species, occurring on plant material and in soil.

Reverse greenish-black

Cladosporium cladosporioides DBM 4145