. The Theoretical Mycology

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Third Class / Biology / Biology / Sciences College

In the world's organisms are divided into five groups:

1-Prokaryota:

a----Eubacteria

b----Archaebacteria

2-Eukaryota:

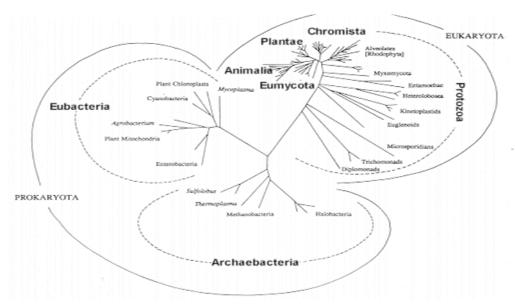
a----Protozoa

b----Chromista(Straminipila)

c---- Eumycota

d----Plantae

E---- Animalia



The general characteristics of fungi:

- 1- Organisms lacking chlorophyll.
- 2-Nutrition by absorption (saprophyte or parasite).
- 3- Fungi body consists of a single cell, such as a yeast or a multi celular-such as molds (mycelium).
- 4- Fungal cell is surrounded by a cell wall, except Myxomycetes.
- 5- Fungi reproduce by forming spores (sexual or asexual) .

Defined fungi as eukaryotic, spore bearing, lacking chlorophyll that may reproduce sexual or asexual and whose filamentous, branched and somatic structures are typically surrounded by cell walls containing chitin, cellulose or both of these substances with many other complex carbohydrates.

With photosynthetic pigments being absent, fungi have a heterotrophic mode of nutrition. In contrast to animals which typically feed by ingestion, fungi obtain their nutrients by extracellular digestion due to the activity of secreted enzymes, followed by absorption of the solubilized breakdown products. The combination of extracellular digestion and absorption can be seen as the ultimate determinant of the fungal lifestyle. often patchy resources is greatly facilitated by the production of numerous small spores .growth as a system of branching tubes(, the hyphae) which together make up the mycelium. Hyphae are generally quite uniform in different taxonomic groups of fungi. One of the few features of distinction that they do offer is the presence or absence of cross-walls or septa. The **Oomycota and Zygomycota** generally have aseptate hyphae in which the nuclei lie in a common mass of cytoplasm. Such a condition is described as coenocytic.

In contrast, **Asco- and Basidiomycota** and their ssociated asexual states generally have septate hyphae in which each segment contains one, two or more nuclei. If the nuclei are genetically identical, as in a mycelium derived from a single uninucleate spore, the mycelium is said to be **homokaryotic**,

but where a cell or mycelium contains nuclei of different genotype, e.g. as a result of fusion (anastomosis) of genetically different hyphae, it is said to be heterokaryotic. A special condition is found in the mycelium of many Basidiomycota in which each cell contains two genetically distinct nuclei. This condition is dikaryotic, to distinguish it from mycelia which are monokaryotic. It should be noted that septa, where present, are usually perforated and allow for the exchange of cytoplasm or organelles. Not all fungi grow as hyphae. Some grow as discrete yeast cells which divide by fission or, more frequently, budding Yeasts are common, especially in situations where efficient penetration of the substratum is not required, on plant surfaces or in the digestive tracts of animals. A few species, including certain pathogens of humans and animals, are dimorphic (capable of switching between hyphal and yeast-like growth forms) Intermediate stages between yeast cells and true hyphae also occur and are termed pseudohyphae. Some lower fungi grow as a thallus, a walled structure in which the protoplasm is concentrated in one or more centres from which root-like branches (rhizoids) ramify Certain obligately pathogenic fungi and fungus-like organisms grow as a naked plasmodium a uni- or multinucleate mass of protoplasm not surrounded by acell wall of its own, or as pseudoplasmodium of amoeboid cells which retain their individual plasma membranes.

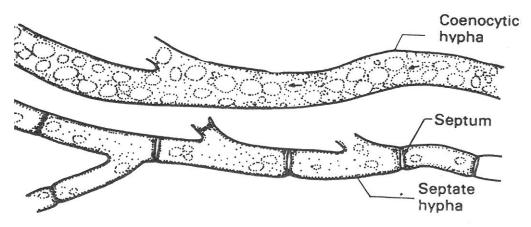


Fig. 1.1: Two types of somatic hyphae.

The fungal cell:

1-fungal cell wall:

a—Microfibrils.(consist of Chitine or Chitosan / Cellulose / Glucans / Protein/ Lipid .(cellulose microfibrils in the Oomycota only but not contane chitine).

b—Matrix.

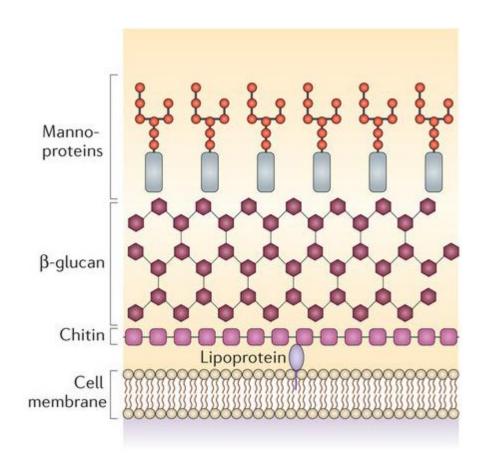
Cell wall components are different depending on Each cell fungal solid wall gives the cell shape and keeps the components from external effects and consists of the cell wall in fungi, mainly from multiple sugars and relatively small amounts of proteins and fats, and inorganic ions. Although the chemical composition of cell walls can vary considerably between and within different groups of fungi ,the basic design seems to be universal. It consists of a structural scaffold of **fibres** which are crosslinked, and a **matrix** of gel-like or crystalline material ,The degree of cross-linking will determine the plasticity (extensibility) of the wall, whereas the pore size (permeability) is a property of the wall matrix.

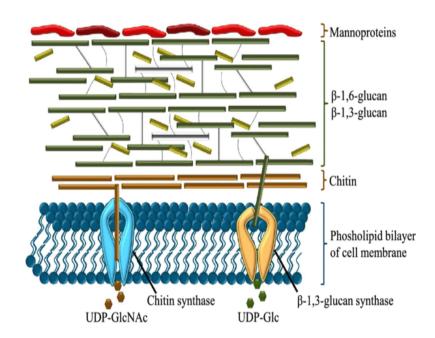
The scaffold forms the inner layer of the wall and the matrix is found predominantly in the outer layer. In the Ascomycota and Basidiomycota, the fibres are chitin microfibrils, i.e. bundles of linear b-(1,4)-linked N-acetylglucosamine chains, In the Zygomycota, the chitin fibres are modified after their synthesis by partial or complete deacetylation to produce poly-b-(1,4)-glucosamine, which is called chitosan fibres are cross-linked by polysaccharides containing glucuronic acid and various neutral sugars, The cell wall matrix comprises glucans and proteins,

Oomycota from the 'true fungi' (Eumycota) has been the absence of chitin from their cell walls even though chitin is now known to be produced by certain species of Oomycota under certain conditions, the structural is filled by cellulose, As in many other fungi, the fibres thus produced are cross-linked by an alkali-insoluble glucan containing b-(1,3)- and b-(1,6)-linkages. In additionto proteins, the main matrix component appears to be an alkali-soluble b-(1,3)-glucan.

fungi, as well as different stages of growth, as well as different environmental factors surrounding it.

The cell wall is composed of several layers as follows:





Group	Example	Chitin	Cellulose	Glucans	Protein	Lipid
Oomycota	Phytophthora	0	25	65	4	2
Chytridiomycota	Allomyces	58	0	16	10	?
Zygomycota	Mucor	9*	0	44	6	8
Ascomycota	Saccharomyces	1	0	60	13	8
	Fusarium	39	0	29	7	6
Basidiomycota	Schizophyllum	5	0	81	2	?
	Coprinus	33	0	50	10	?

^{*}Mainly chitosan.

Structural formulae of the principal fibrous components of fungal :

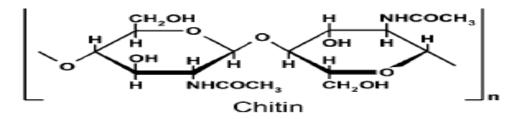
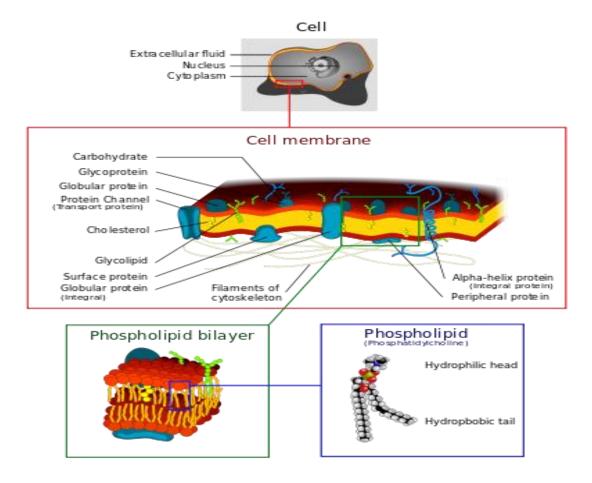


Fig 1.5 Structural formulae of the principal fibrous components of fungal cell walls.

The cell membrane:

is a biological membrane that separates the interior of all cells from the outside environment. The cell membrane is selectively permeable to ions and organic molecules and controls the movement of substances in and out of cells. (A semipermeable membrane, also termed a selectively permeable membrane, a partially permeable membrane or a differentially permeable membrane, is a membrane that will allow certain molecules or ions to pass through it by diffusion and occasionally specialized "facilitated diffusion").1 The basic function of the cell membrane is to protect the cell from its surroundings. It consists of the lipid bilayer with embedded proteins. Cell membranes are involved in a variety of cellular processes such as cell adhesion, ion conductivity and cell signaling and serve as the attachment surface for several extracellular structures, including the cell wall, glycocalyx, and intracellular cytoskeleton. Cell membranes can be artificially reassembled.



The endoplasmic reticulum (ER):

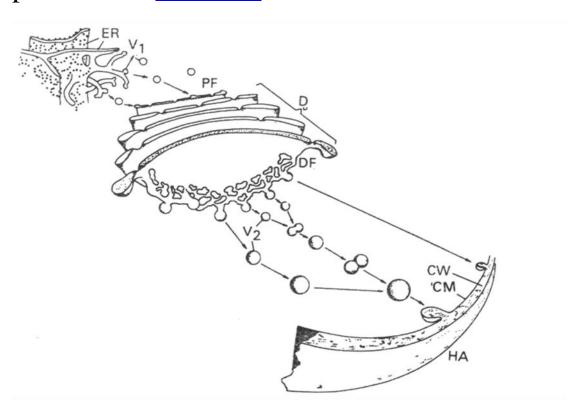
is an <u>organelle</u> of <u>cells</u> in <u>eukaryotic organisms</u> that forms an interconnected network of membrane vesicles. According to the structure the endoplasmic reticulum is classified into two types, that is, **rough endoplasmic reticulum**[[] (**RER**) and **smooth endoplasmic reticulum** (**SER**).

The rough endoplasmic reticulum is studded with ribosomes on the cytosolic face. These are the sites of protein synthesis.

The smooth endoplasmic reticulum is concerned with *lipid metabolism*, *carbohydrate metabolism* and *detoxification*.

The Golgi apparatus:

Part of the cellular <u>endomembrane system</u>, the Golgi apparatus packages proteins inside the cell before they are sent to their destination; it is particularly important in the processing of proteins for <u>secretion</u>.



The material is transferred from the endoplasmic reticulum (ER) to the dictyosome(D) by blebbing of the endoplasmic reticulum and

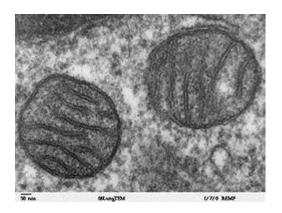
coalescene of vesicles (V) to forma cisterna at the proximal face (P F) of the dictyosome. The contents of the cisterna and membranes are then transformed as the cisterna is moved to the distal face (D F) of the dictyosome. Secretory vesicle (V) released from the cisterna enlarge and / or fuse with one another , Migrate to the apex of the hypha and fuse with cell membrane (C M) liberating their contents to the cell wall (C W) .

Nucleus:

The nucleus is a highly specialized organelle that serves as the information and administrative center of the cell. This organelle has two major functions. It stores the cell's hereditary material, or DNA, and it coordinates the cell's activities, which include intermediary metabolism, growth, protein synthesis, and reproduction (cell division).

Mitochondrion:

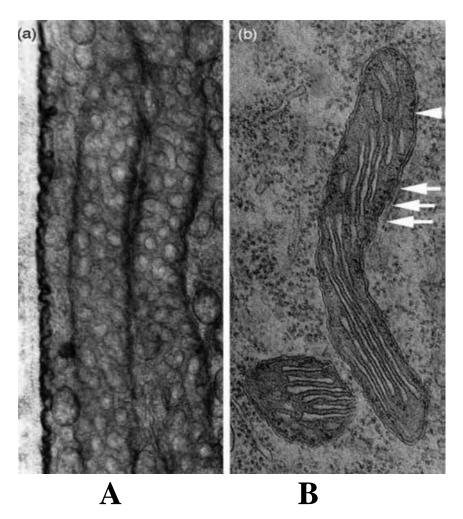
"cellular power plants" because they generate most of the cell's supply of <u>adenosine triphosphate</u> (ATP), used as a source of <u>chemical energy</u> In addition to supplying cellular energy



The number of mitochondria in a cell varies widely by <u>organism</u> and <u>tissue</u> type. Many cells have only a single mitochondrion, whereas others can contain several thousand mitochondria. The organelle is composed of compartments that carry out specialized functions. These compartments or regions include the <u>outer membrane</u>, 12 the <u>intermembrane space</u>, the <u>inner membrane</u>, and the <u>cristae</u> and <u>matrix</u>.

Mitochondrial ultrastructure observed by transmission electronmicroscopy.

- (a) Mitochondrion of *Phytophthora erythroseptica* (Oomycota) (all Straminipilla). The inner Mitochondrial membrane is folded into a complex tubular network.
- **(b)** Mitochondrion of *Sordaria fimicola* (Ascomycota) (all Eumycota) with the inner membrane appearing lamellate. Mitochondrial.



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Classification of fungi

The scientists studied fungi noted the variety and differences in fungi and to facilitate their studies classify it . Persoon (1801) has the first put classification of fungi. Years ago fungi classified as according to the following:

Superkingdom: Eukaryota

Kingdom: Myceteae

Division:

1-Gymnomycota .(Myxomycetes)

2-Mastigomycota . subdivision : a-Haplomastigomycotina . b-Diplomastigomycotina .

- 3-Amastigomycota.
- 1-Zygomycotina. 2-Ascomycotina.
- 3- Basidiomycotina. 4-Deuteromycotina.

But at the present time class scientists fungi, depending on the external appearance and internal structure and genetic and the presence flagella and it type and tracks the biochemistry .Class Fungi as the following:

1-KINGDOM: PROTOZOA

1-Phylum: Myxomycota: **(1)**

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Acrasiomycetes

Dictyosteliomycetes

Protosteliomycetes

Myxomycetes

2- Phylum: Plasmodiophoromycota:

Plasmodiophorales

Haptoglossales (Oomycota?)

2-KINGDOM: STRAMINIPILA

1-Phylum: Hyphochytriomycota

2-Phylum: Labyrinthulomycota

Labyrinthulomycetes

Thraustochytriomycetes

3-Phylum: Oomycota

Saprolegniales

Pythiales

Peronosporales

3-KINGDOM: FUNGI (EUMYCOTA)

1-phylum: Chytridiomycota: Chytridiomycetes

2-phylum: Zygomycota:

a-Zygomycetes b-Trichomycetes

3-Phylum: Ascomycota

Archiascomycetes

Hemiascomycetes

Plectomycetes

Hymenoascomycetes

Pyrenomycetes

Erysiphales

Pezizales

Helotiales

Lecanorales/lichens

(2)

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Loculoascomycetes

4-Phylum: Basidiomycota

Homobasidiomycetes

Homobasidiomycetes: gasteromycetes

Heterobasidiomycetes

Urediniomycetes

Ustilaginomycetes

. Example of the hierarchy of taxonomic terms. The wheat stem rust fungus, Puccinia graminis, is used as an example :.

Kingdom: Fungi

Subkingdom : Eumycota

Phylum: Basidiomycota Class: Urediniomycetes

Order : Uredinales Family : Pucciniaceae

Genus : Puccinia Species : graminis Puccinia graminis

Kingdom: Fungi

Division: Ascomycota

Class: <u>Eurotiomycetes</u>

Order: <u>Eurotiales</u>

Family: <u>Trichocomaceae</u>

Genus: Aspergillus

Species: niger

A. niger

Lect.4:

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What are fungi?

It is too hard to give a constant definition of fungi due to the great number of this group, their diversified and overlapped characteristics. Still, the term fungi is referred to the group of the organisms that are characterized by having Eukaryote and Spores-bearing (Spores = germs), non-chlorophyll, sexual and asexual reproduction and their bodies are usually formed of varied linear structures and their cells are surrounded with a cellular wall .These walls contain Chitin .

General characters of fungi

Fungi are the eukaryotic, achlorophyllous, and unicellular or multicellular organisms, which may reproduce by asexual and sexual spores.

- 1. All are eukaryotic Possess membrane-bound nuclei (containing chromosomes) and a range of membrane-bound cytoplasmic organelles (e.g. mitochondria, vacuoles, endoplasmic reticulum).
- 2. Most are filamentous Composed of individual microscopic filaments called hyphae, which exhibit apical growth and which branch to form a network of hyphae called a mycelium.
- 3. Some are unicellular e.g. yeasts.
- 4. Protoplasm of a hypha or cell is surrounded by a rigid wall Composed primarily of chitin and glucans, although the walls of some species contain cellulose.

- 5. Many reproduce both sexually and asexually Both sexual and asexual reproduction often result in the production of spores.
- 6. Their nuclei are typically haploid and hyphal compartments are often multinucleate Although the oomycota and some yeast possess diploid nuclei.
- 7. All are achlorophyllous They lack chlorophyll pigments and are incapable of photosynthesis.
- 8. All are chemoheterotrophic (chemo-organotrophic) They utilise preexisting organic sources of carbon in their environment and the energy from chemical reactions to synthesize the organic compounds they require for growth and energy.
- 9. Possess characteristic range of storage compounds e.g. trehalose, glycogen, sugar alcohols and lipids.
- 10. May be free-living or may form intimate relationships with other organisms i.e. may be freeliving, parasitic or mutualistic (symbiotic).

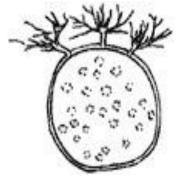
Structure of fungal body

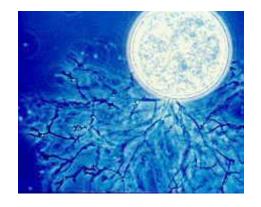
Thallus:

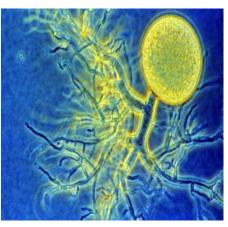
The body of the fungus is called as 'thallus'.

Eucarpic thallus:

The thallus is differentiated into vegetative part, which absorbs nutrients, and a reproductive part, which forms reproductive structure. Such thalli are called as eucarpic. e.g. Pythium aphanidermatum.

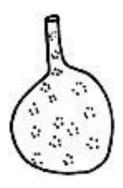






Holocarpic thallus:

The thallus does not show any differentiation on vegetative and reproductive structure. After a phase of vegetative growth, it gets converted into one or more reproductive structures. Such thalli are called as 'holocarpic' e.g. yeast, Synchytrium endobioticum .

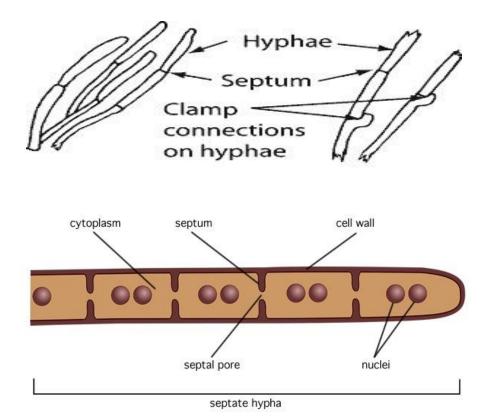




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Hyphae

Hyphae is a tubular, transparent filament, usually branched, composed of an outer cell wall and a cavity (lumen) lined or filled with protoplasm including cytoplasm. Hyphae are divided into compartments or cells by cross walls called septa and are generally called as septate (with cross wall) or coenocytic (aseptate -without cross wall). Hyphae of most of the fungi measure 5-10 μm .



Mycelium (pl. Mycelia):

The hyphal mass or network of hyphae constituting the body (thallus) of the fungus is called as mycelium. The mycelium of parasitic fungi grows on the surface of the host and spread between the cells and it is called intercellular mycelium. The mycelium of parasitic fungi, which

grows on the surface of the host and penetrates into the host cells and is called intracellular mycelium. If the mycelium is intercellular, food is absorbed through the host cell walls or membrane. If the mycelium penetrates into the cells, the hyphal walls come into direct contact with the host protoplasm. Intercellular hyphae of many fungi, especially of obligate parasites of plants (fungi causing downy mildews, powdery mildews and rusts) obtain nutrients through haustoria.

Monokaryotic mycelium (uninucleate): Mycelium contains single nucleus that usually forms part of haplophase in the life cycle of fungi.

Dikaryotic mycelium (binucleate): Mycelium contains pair of nuclei (dikaryon), which denotes the diplophase in the life cycle of fungi.



Homokaryotic mycelium: The mycelium contains genetically identical nuclei.

Heterokaryotic mycelium: The mycelium contains nuclei of different genetic constituents.

Multinucleate: The fungal cell contains more than 2 nuclei.

Lect.5:

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Septa

Transverse septa occur in the thallus of all filamentous fungi to cut off reproductive cells from the rest of the hypha, to separate off the damaged parts or to divide the hypha into regular or irregular compartments or cells. There are two general types of septa in fungi viz., primary and adventitious. The primary septa are formed in association with nuclear division and are laid down between daughter nuclei. The adventitious septa are formed independently of nuclear division and are especially associated with changes in the concentration of the protoplasm as it moves from one part of the hypha to another.

Transverse septa

Septa vary in their construction septa have biological importance in the lifecycle of fungi. Some are simple whereas others are complex. All types of septa are formed by centripetal growth from the hyphal wall inward. In some septa, the growth continues until the septum is a solid plate. In others the septum remains incomplete, leaving a pore in the centre that may often be plugged or occulted. Some groups of Basidiomycetes like Auriculariaceae, Tremellaceae, Aphyllophorales, Agaricales etc (except Ustilaginales and uredinales) have more complex septa. Surrounding the central pore in the septum is a curved flange of wall material, which is thickened to form a barrel-shaped or cylindrical structure surrounding the pore. Septa of this type are termed dolipore septa (L. dolium = a large jar or cask i . e., barrel).

These septa are often overlaid by perforated cap, which is an extension of the endoplasmic reticulum. This cap is known as parenthosome or pore cap. Despite these apparent barriers, there is a good cytoplasmic continuity between adjacent cells. The septal pore may vary in width from 0.1 to 0.2 μm . Dolipore septa are found in both monokaryotic and dikaryotic mycelia.

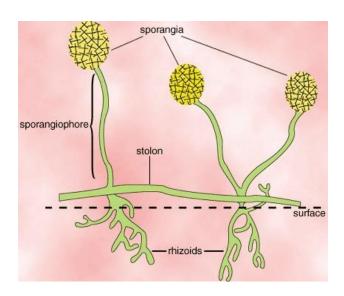
Fungal cell structure

Fungal cells are typically eukaryotic and have distinguished characteristics than that of bacteria, and algae. The chief components of cell wall appears to be various types of carbohydrate or their mixtures (upto 80-90%) such as cellulose, pectose, callose etc., cellulose predominates in the cell wall of mastigomycotina (lower fungi) while in higher fungi chitin is present. The living protoplast of the fungal cell is enclosed in a cell membrane called as plasma membrane or plasmalemma. Cytoplasm contains organelles such nucleus. mitochondria, ribosomes, Golgi apparatus, vacuoles, vesicles. microbodies, endoplasmic reticulum, lysosomes and microtubules.

The fungal nucleus has nuclear envelope comprising of two typical unit membrane and a central dense area known as nucleolus, which mainly consist of RNA. In multinucleate hyphae, the nuclei may be interconnected by the endoplasmic reticulum. Vacuoles present inside the cell provide turgor needed for cell growth and maintenance of cell shape. Beside the osmotic function, they also store reserve materials. The chief storage products of fungi are glycogen and lipid. The apex of the hyphae are usually rich in vesicles and are called as apical vesicular complex (AVC) which helps in the transportation of products formed by the secretary action of golgi apparatus to the site where these products are utilized.

Specialized Somatic Structures

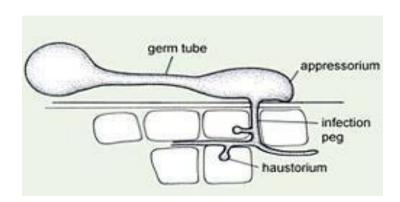
Rhizoid A rhizoid (Gr. rhiza = root + oeides = like) is a short, root-like filamentous outgrowth of the thallus generally formed in tufts at the base of small unicellular thalli or small porophores. Rhizoid serves as anchoring or attachment organ to the substratum and also as an organ of absorption of nutrients from substratum. Rhizoids are short, delicate filaments that contain protoplasm but no nuclei.



Rhizoids are common in lower fungi like Chytridiomycetes, Oomycetes and Zygomycetes. Some species produce a many-branched rhizomycelium. This is an extensive rhizoidal system that usually do not contains nuclei, but through which nuclei migrate. e.g. Cladochytrium sp. On rhizomycelium numerous sporangia develop. Such thalli are polycentric, that is, they form several reproductive centres instead of a single one where the thallus is termed monocentric.

Appressorium

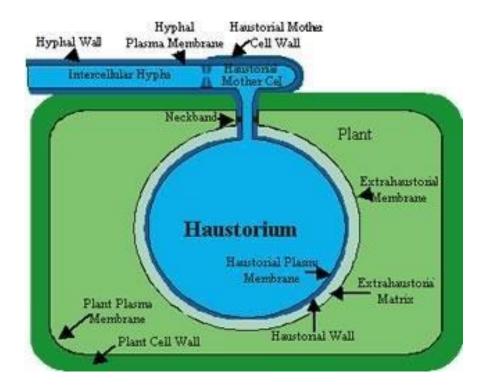
Appressorium (p1. appressorium; L. apprimere = to press against) is a simple or lobed structure of hyphal or germ tube and a pressing organ from which a minute infection peg usually grow and enter the epidermal cell of the host. It helps germ tube or hypha to attach to the surface of the host or substrates. These appressoria are formed from germ tubes of Uredinales (rust fungi), Erysiphales (powdery mildew fungi) and other fungi in their parasitic or saprophytic stages. In addition to giving anchorage, appressoria help the penetrating hyphae, branches to pierce the host cuticle. In fungi like Colletotrichum falcatum, germ tubes from conidia and resulting hyphae form appressoria on coming in contact with any hard surface like soil etc. These appressoria are thought to function as resting structures (chlamydospores) also.



Haustoria

Haustoria (sing. haustorium; L. haustor = drinker) are special hyphal structures or outgrowths of somatic hyphae sent into the cell to absorb nutrients. The hyphal branch said to function as haustorium becomes extremely thin and pointed while piercing the hast cell wall and expands in the cell cavity to form a wider, simple or branched haustorium. Haustoria may be knob-like or balloon – like in shape, elongated or

branched like a miniature root system. The hyphae of obligate parasites of plants like downy mildew, powdery mildew or rust fungi late blight fungus etc., produce haustoria. Hyphopodia: Hyphopodium (pl. hyphopodia Gr. hyphe = web + pous = foot) is a small appendage with one or two cells in length on an external hypha and function as absorbing structures. The terminal cell of hyphopodium is expanded and rounded or pointed. Sometimes it produces a haustorium. e.g. Ectophytic fungi (Meliola aesariae) attacking leaves of green plants.



Lect.6:

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Aggregations of hyphae and tissues

a. Mycelial strand

Mycelial strands aggregates of parallel are undifferentiated hyphae, which adhere closely and are frequently anastomosed or cemented together. They are relatively loose (e.g. Sclerotium rolfsii growth on culture medium) compared to rhizomorph. They have no welldefined apical meristem. Mycelial strand formation is quite common in Basidiomycetes, Ascomycetes and Deuteromycetes. Mycelial strands form the familiar 'spawn' of the cultivated mushroom, Agaricus bisporus. Mycelial strands are capable of translocating materials in both the directions. They are believed to afford means by which a fungus can extend an established food base and colonize a new substratum, by increasing the inoculum potential of the fungus at the point of colonization.

b. Rhizomorph

Rhizomorph (Gr. rhiza=root + morphe = shape) is the aggregation of highly differentiated hyphae with a well defined apical meristem, a central core of larger, thin walled, cells which are often darkly pigmented. These root-like aggregation is found in the honey fungus or honey agaric Armillariella mellea (=Armillaria mellea). They grow faster than the mycelial strands. The growing tip of rhizomorph resembles that of a root

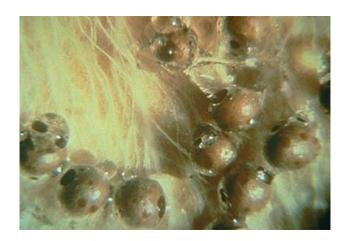
tip. The fungus may spread underground from one root system to another by means of rhizomorph .

c. Fungal tissues

During certain stages of the life cycle of most fungi, the mycelium becomes organized into loosely or compactly woven tissues. These organized fungal tissues are called plectenchyma (Gr. plekein = to weave + encyma = infusion i.e., a woven tissue). There are two types of plectenchyma viz., prosenchyma and pseudoparenchyma. When the tissue is loosely woven and the hyphae lie parallel to one another it is called prosenchyma (Gr. pros = toward + enchyma = infusion, i.e., approaching a tissue). These tissues have distinguishable and typical elongated cells. Pseudoparenchyma (Gr. Pseudo = false) consists of closely packed, more or less isodiametric or oval cells resembling the parenchyma cells of vascular plants. In this type of tissues hyphae lose their individuality and are not distinguishable. Cells in prosenchyma are thinwalled and cells in pseudoparenchyma.

* Stroma and Sclerotium

I: Stromata and sclerotia are somatic structures of fungi. i. Stroma (pl. stromata; Gr. stroma = mattress) A stroma is a compact, somatic structure or hyphal aggregation similar to a mattress or a cushion, on which or in which fructifications of fungi are usually formed. They may be of various shapes and sizes. Hyphal masses like acervuli, sporodochia, pionnotes etc. are the fertile stromata, which bear sporophores producing spores. ii. II: Sclerotium (pl. sclerotia; Gr. skeleros = hard) A sclerotium is a resting body formed by aggregation of somatic hyphae into dense, rounded, flattened, elongated or horn-shaped dark masses.



They are thick-walled resting structures, which contain food reserves. Sclerotia are hard structures resistant to unfavourable physical and chemical conditions. They may remain dormant for longer periods of time, sometimes for several years and germinate on the return of favourable conditions. The sclerotia on germination may be myceliogenous and produce directly the mycelium e.g. *Sclerotium rolfsii, Rhizoctonia solani* and *S. cepivorum* (white rot of onion).

They may be sporogenous and bear mass of spores. e.g. *Botrytis cinerea*. They may also be carpogenous where in they produce a spore fruit (ascocarps or basidiocarps) bearing stalk. e.g. Sclerotinia sp. Claviceps purpurea (ergot of rye). Development of ascocarps is seen in Sclerotinia, where stalked cups or apothecia, bearing asci, arise from sclerotia. In *Claviceps purpurea, sclerotia germinate* and give rise to drumstick like structures called perithecial stromata, which contain perithecia, flask-shaped cavities within which the asci are formed.

Mycorrhizae

Mycorrhiza (pl. mycorrhizae; Gr. mykes = mushroom + rhiza = root) is the symbiotic association between higher plant roots and fungal mycelia. Many plants in nature have mycorrhizal associations.

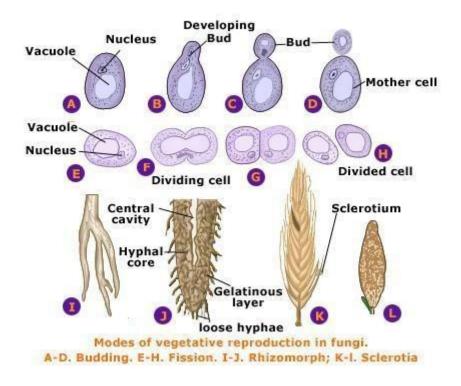
Mycorrhizal plants increase the surface area of the root system for better absorption of nutrients from soil especially when the soils are deficient in phosphorus. The nature of association is believed to be symbiotic (mutualism), non-pathogenic or weakly pathogenic. There are three types of mycorrhizal fungal associations with plant roots. They are ectotrophic or sheathing or ectomycorrhiza, endotrophic or endomycorrhiza and ectendotrophicmycorrhiza

Reproduction

Reproduction is the formation of new individuals having all the characteristics typical of a species. The fungi reproduce by means of asexual and sexual or parasexual reproduction. Asexual reproduction is sometimes called somatic or vegetative and it does not involve union of nuclei, sex cells or sex organs. The union of two nuclei characterizes sexual reproduction.

Asexual Reproduction

In fungi, asexual reproduction is more important for the propagation of species. Asexual reproduction does not involve union of sex organs (gametangia) or sex cells (gametes) or nuclei. In fungi the following are the common methods of asexual reproduction.



1. Fragmentation of mycelium

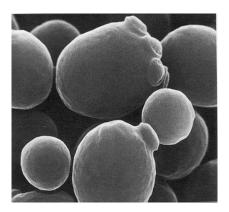
Mycelial fragments from any part of the thallus may grow into new individuals when suitable conditions are provided.

2. Fission of unicellular thalli

It is also known as transverse cell division. Reproduction by the method of fission is are in fungi. Fission is simple splitting of cells into two daughter cells by constriction and the formation of a cell wall. It is observed in *Schizosaccharomyces* spp.

3. **Budding**

Budding is the production of a small outgrowth (bud) from a parent cell. As the bud is formed, the nucleus of the parent cell divides and one daughter nucleus migrates into the bud. The bud increases in size, while still attached to the parent cell and eventually breaks off and forms a new individual. It is common in yeasts.(*Saccharomyces* sp.).



Scanning electron micrograph of the budding yeast Saccharomyces erevisiae.

4. Production of asexual spores Reproduction by the production of spores is very common in many fungi.

Lect.9:

Dr. Warka Saeed Qassim Al-Taee

SPORES

The term 'spore'(Gr. spora=seed, spore) is applied to any small propagative, reproductive or survival unit, which separates from a hypha or sporogenous cell and can grow independently into a new individual. Spores may be unicellular or multicellular. Multicellular spores are mostly with transverse septa and in some genera like Alternaria a spore will have both transverse and longitudinal septa. Each cell of a multicellular spore may be uninucleate, binucleate or multinucleate depending on the fungal species. The spores may be in different shapes and sizes.

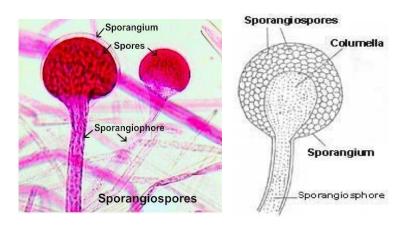
They may be spherical, oval or ovate, obovate, pyriform, obpyriform, ellipsoid, cylindrical, oblong, allantoid, filiform or selecoid, falcate or fusion. The spores may be with or without simple or branched appendages. The spores may be motile or nonmotile. If the spores are motile they are called planospores (Gr. Planets = wanderer) and nonmotile spores are called aplanospores. Spores may be thin or thickwalled, hyaline or coloured, smooth or with ornamented walls. The following types of ornamentations are found on the walls.

Asexual spores

The spores produced asexual means are:

- a. Sporangiospores
- b. Conidia
- c. Chlamydospores

a. Sporangiospores: may be motile (planospores) or nonmotile spores (aplanospores). In simpler fungi sporangiospores are usually motile and are called zoospores. These spores are produced in lower fungi, which inhabit aquatic or moist terrestrial substrates. sporangiospores are formed in globose or sac-like structure called sporangium (pl. sporangia; Gr. Spora = seed, spore + angeion = vessel). In the zygomycetes and especially in the 30 Plant Pathogens & Principles of Plant Pathology www.AgriMoon.Com Mucorales, the non-motile asexual spores called aplanospores are contained in globose sporangia surrounding a central core or columella. Sporangia are also known in which there is no columella, or where the spores (aplanospores) are arranged in a row inside a cylindrical sac termed a Merosporangium (e.g. Syncephalastrum spp.Mucorales).



These aplanospores may be uni or multinucleate and are unicellular, generally smoothwalled, globose or ellipsoid in shape. When aplanospores mature, they may be surrounded by mucilage and rain splash or insects usually disperse such spores. When aplanospores are dry then are dispersed by wind currents. The sporangiospores for sporangium may vary from several thousands to only one. In some fungi few-spores sporangia are called Sporangiola. Sporangiola are dispersed as a unit. e.g. Choanephora sp. and Blakeslee sp. in Choanephoraceae of Mucorales. In holocarpic thalli, the entire thallus

(without differentiation of a sporophore) becomes a sporangium. Its contents cleave into a number of segments which round off and become zoospores. In eucarpic thalli, a part of the thallus, or special branches from thallus, function as or produce sporangia.

It is an asexually produced spore, which is motile by means of flagellum or flagella. Zoospore is naked and its covering is only a hyaloplasm membrane. Normally, zoospores are uninucleate and haploid. Zoospores may be spherical, oval, pyriform, obpyriform, elongate or reniform in shape. The zoospores are provided with one or two flagella (sing. flagellum, L. flagellum=whip) for its movement in the surrounding film of water. Flagellum is a hair-or tinsellike structure that serves to propel a motile cell.

These flagella may be anterior, posterior or laterally attached to a groove in the body. There are two types of flagella in zoospores. They are whiplash and tinsel types. The whiplash flagellum has a long rigid base composed of all the eleven fibrils and a short flexible end formed of the two central fibrils only. The tinsel flagellum has a rachis, which is covered on all sides along its centre length with short fibrils. In uniflagellate zoospores the flagellum may be anterior.

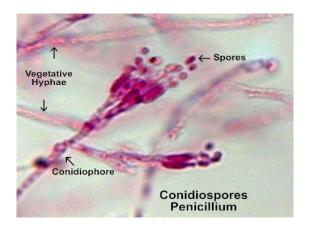
or posterior. But in biflagellate zoospores one is whiplash and the other is tinsel type and one points forward and the other backward. But in Plasmodiophorales fungi flagella are of whiplash type and unequal.

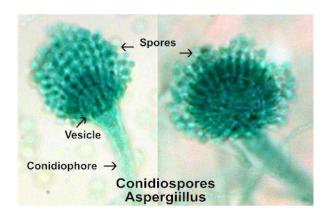
Zoospores pass through the three phases viz., motility, encasement and germination. The length of their motility depends on available moisture, temperature and presence of stimulatory or inhibitory

substances in the environment. Later the zoospores become sluggish, spend or cast their flagella (except in chytridiacious fungi and primary zoospores in Saprolegniales where flagella are shed but withdrawn into its body become spherical and secrete thin wall around itself and become encysted. The encysted zoospores germinate. The functions of zoospores include initiation of new generation and acting as gametes.

b. Conidiospores Conidiospores or conidia (sing. Conidium)

are asexual reproductive structures borne on special spore bearing hyphae conidiophores. They are found in many different groups of fungi, but especially in ascomycotina, Basidiomycotina and Deuteromycotina. In Deuteromycotina conidia are the only means of reproduction. Conidia may be borne singly or in chains or in cluster. They vary from unicellular (e.g. Colletotrichum), bicellular, microconidia of Fusarium spp. and multicellular (Pestalotiopsis, Cercospora). One-celled spores are called amerospores, two celled spores are didymospores and multicellular spores are called phragmospores. The multicellular conidia may be divided by the septa in one to three planes. In Alternaria spp., conidia are with both transverse and longitudinal septa are called dictyospores.



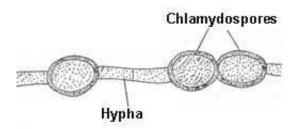


The shape of the conidium may vary. They may be globose, elliptical, ovoid, cylindrical, branched or spirally coiled or star-shaped (staurospores). The colour of the conidia may be 32 Plant Pathogens & Principles of Plant Pathology www.AgriMoon.Com hyaline (hyalospore) or coloured (phaeospore) pink, green, or dark. The dark pigments are probably melanins. The colour of the conidia and conidiophores are important features used in classification. In the order Entomophthorales (e.g. Basidiobolus, Pilobolus) asexual reproduction is by means or forcibly discharged uninucleate or multinucleate primary conidia. On germination primary conidia develops uninucleate or binucleate secondary conidia. In species of Fusarium one or two-celled microconidia and many-celled macroconidia are common.

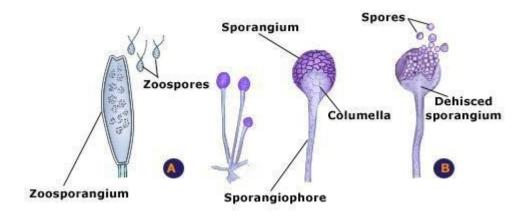
Conidia may be formed in acropetal (oldest conidium at the base and the youngest at the apex) or basipetal (oldest conidium at the apex and youngest at the base) succession. Generally the term 'conidia'is used for any asexual spores other than sporangia and spores formed directly by hyphal cells. When the spore is not much differentiated from the cells of the conidiophore in shape the term oidium is often used for conidia. A distinction between sporangiospores and conidia is that, before germination of sporangiospores a new wall, eventually continuous with the germ tube, is laid down within the original spore wall whilst in conidia there is no new wall layer laid down. Conidiophores are also known as sporophores. They are special hyphae bearing conidia. They may be free, simple or branched.

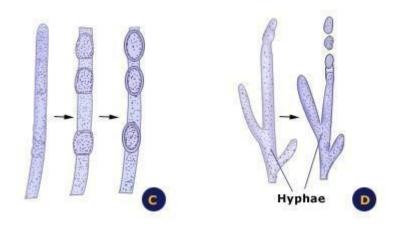
c. Chlamydospores

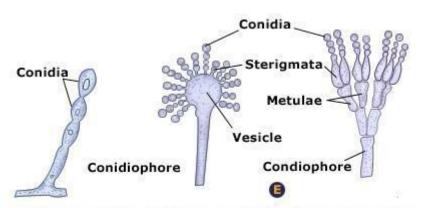
Chlamydospore (Gr. Chlamys = mantle + spora = seed, spore) is a thick-walled thallic conidium that generally function as a resting spore. Terminal or intercalary segments or mycelium may become packed with food reserves and develop thick walls. The walls may be colourless or pigmented with dark melanin pigment.



These structures are known as chlamydospores. e.g. Fusarium, Mucor racemosus, Saprolegnia. Generally there is no mechanism for detachment and dispersal of chlamydospores. They become separated from each other by the disintegration of intervening hyphae. They are the important organs or asexual survival in soil fungi. When chlamydospores are found in between fungal cells they are called 'intercalary chlamydospores'. Chlamydospores produced at the apex of the hypha are called 'apical or terminal chlamydospores'







A. Zoospores; B. Sporangiospores; C. Chalmydospores; D. Oidia; E. Conidia

Lect .10:

Dr. Warka Saeed Qassim Al-Taee

Sexual Reproduction

Sexual reproduction in fungi involves union of two compatible nuclei. The nuclei may be carried in motile or non-motile gametes, in gametangia or in somatic cells of the thallus

Phases of sexual reproduction

Three typical phases occur in sequence during the sexual reproduction.

1. Plasmogamy

In plasmogamy (Gr. plasma=a molded object, i.e. a being + gamos = marriage, union) anastomosis of two cells or gametes and fusion of their protoplasts take place. In the process the two haploid nuclei of opposite sexes (compatible nuclei) are brought together but eh nuclei will not fuse.

2. Karyogamy

The fusion of two haploid nuclei brought together as a result of plasmogamy is called karyogamy (Gr. karyon = nut, nucleus + gamos = marriage). This stage follows immediately after plasmogamy in many of the lower fungi or may be delayed in higher fungi. In higher fungi plasmogamy results in a binucleate cell containing one nucleus from each cell. Such a pair of nuclei is called dikaryon(NL. Di = two + Gr. karyon = nut). These two nuclei may not fuse until later in the life history of the fungus. Meanwhile, during growth and cell division of the binucleate cell, the dikaryotic condition may be perpetuated from cell to cell by conjugate division of the two closely associated nuclei and by the separation of the resulting sister nuclei with two

daughter cells. Nuclear fusion, which eventually takes place in all sexually reproducing fungi, is followed by meiosis.

3. Meiosis

Karyogamy results in the formation of a diploid (2n) nucleus. Meiosis (Gr. meiosis=reduction) reduces the number of chromosomes to haploid and constitutes the third phase of the sexual reproduction. This nucleus undergoes a reduction division to form two haploid nuclei each with 'n'chromosomes. A mitotic division follows and four nuclei are formed. In ascomycetes another nuclear division takes place resulting in the formation eight nuclei. The nuclei get surrounded by a small amount of cytoplasm and secrete a wall to become spores.

In a true sexual cycle, the above three phases occur in a regular sequence and usually at specified points. If there is only one free living thallus, haploid or diploid in the life cycle of a fungus is called haplobiontic (Gr. haplos = single + bios = life). e.g. Oomycetes haploid gamete and diploid mycelium. If a haploid thallus alternates with a diploid, the life cycle is called diplobiontic (Gr. diplos = double + bios = life). e.g. Allomyces (water mold Coelomomyces, mosquito parasite) and in some yeasts.

Organs involved in sexual reproduction

The sex organs of fungi are called gametangia (sing. gametangium; Gr. gametes = husband + angeion = vessel, container). Sex cells are called gametes and the mother cells (sex organs) are called gametangia. If the gametes and gametangia produced are morphologically identical or similar they are called as isogametes (Gr. ison = equal) and isogametangia respectively. When the gametes and gametangia produced differ in size and structure (morphologically

different) they are called heterogametes (Gr. heteros = other, different) and heterogametangia respectively. In the latter case, the male gametangium is called antheridium (pl. antheridia; Gr. antheros = flowery + idion, dimin. suffix) and the female gametangium is called oogonium (pl. oogonia; Gr. oon = egg + gonos = offspring). The male gamete is known as antherozoid or sperm and the female as an egg or oosphere .

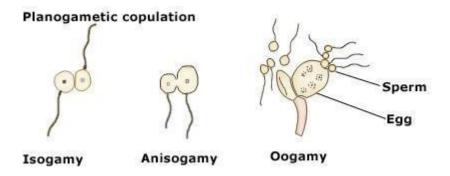
Methods of sexual reproduction

The following are the five methods, which the fungi employ to bring the compatible nuclei together for fusion.

- 1. Planogametic copulation
- 2. Gametangial contact (Gametangy)
- 3. Gametangial copulation (Gametangiogamy)
- 4. Spermatization
- 5. Somatogamy

1. Planogametic copulation

Planogametic copulation or conjugation A planogamate is a motile gamete or sex cell. The fusion of two gametes, one or both of which are motile is called planogametic copulation. This type of sexual reproduction is common in aquatic fungi. There are three different types of planogametic copulation.



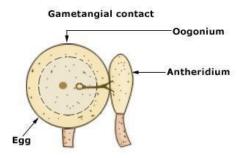
- a. Copulation of isogamous motile gametes In this type morphologically similar butcompatible type of mating type of gametes unite to form a motile zygote. e.g. Synchytrium.
- b. Copulation in anisogamous motile gametes It involves union of one larger gamete with another smaller gamete. The resultant zygote is motile. The zygote resulting from isogamous or anisogamous planogametic copulation forms a 'resting sporangium'. On further development it functions as sporangium by differentiating zoospores internally.
- c. Heterogamous planogametic copulation In this type, a non-motile female gamete (oosphere) is fertilized by a motile male gamete. This results in the formation of oospores, a resistant structure and resting spore. Oospores germinate and produce mycelium directly.

Lect.11:

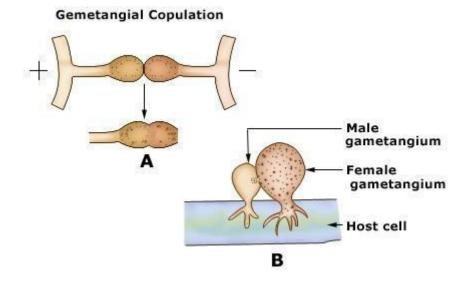
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2. Gametangial contact

In this method the male gamete (antheridium) and the female gamete (oogonium) come in contact and one or more nuclei from the male gamete enter the female gamete, oogonium dissolved in the intervening wall through a pore or through a fertilization tube. In no case the gametangia actually fuse or lose their identify during the sexual act. e.g. Fungi in Peronosporales. Gametangial contact is also common in some Ascomycotina where antheridia and female organs (archigonia or ascogonia) may or may not be well defined.



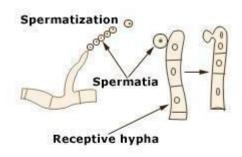
3. Gametangial copulation: This is a process of fusion of entire contents of the two mating gametangia. There are two types. a. Mixing of entire protoplasm of male and female gametangia Two gametangia meet and their entire contents fuse in the female gametangium leading to formation of a zygote. The zygote forms a resting sporangium. e.g. Aquatic fungi (Chytridiomycetes).

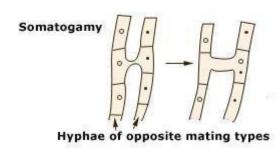


b. Isogamous copulation

Two morphologically similar gametangial hyphae come in contact, the wall at the point of contact dissolves and the contents mix in the cell thus formed. This results in the formation of zygospore. e.g. Mucor, Rhizopus, Phycomyces.

4. Spermatization :Some fungi like rusts bear numerous minute, non-motile uninucleate, male cells called spermatia. (sing. spermatium; Gr. spermation=little seed) Spermatia are produced in spiral receptacles called spermagonia (sing. spermagonium; Gr. Sperma =seed, sperm+ gennao=I give birth) or pycnia (sing. pycnium; Gr. pycnos=concentrated). Insects, wind or water to the female gametangium carries them, which is usually a special receptive hypha (or trichogyne) to which they become attached. A pore develops at the point of contact and the contents of spermatium pass into the particular respective hyphae. This results in plasmogamy and initiation of the dikaryotic stage of the cell.





5. Somatogamy

In somatogamy to sex organs are produced and somatic cells function as gametes somatogamy (Gr. soma = body + gamos = marriage, union) hyphae anastomose and the nuclei of opposite matting type are brought together in one cell. Somatogamy is common in Ascomycotina and Basidiomycotina fungi.

Heterokaryosis

The phenomenon of existence of different kinds of nuclei in the same individual is known as heterokaryosis. (Gr. heteros = other+ karyon=nut, nucleus). The individual which exhibit heterokaryosis is called heterokaryon or heterokaryotic. It has been demonstrated in numerous Ascomycetes, Basidiomycetes and Fungi Imperfecti (Davis, 1966). In a heterokaryotic individual, each nucleus is independent of all other nuclei, but the structure and behaviour of the individual appear to be controlled by the kinds of genes it contains and the proportion of each kind. Heterokaryosis may arise in a fungal thallus in four ways:

- 1. By the germination of a heterokaryotic spore, which will give rise to a heterokaryotic soma
- 2. By the introduction of genetically different nuclei into homokaryon (Gr. homo=same + karyon = nut, nucleus), a soma in which all nuclei are similar.
- 3. By mutation, in a multinucleate homokaryon. The mutant nuclei subsequently survive, multiply and spread among the wild-type nuclei.
- 4. By fusion of some nuclei in a haploid homokaryon to form diploid nuclei which subsequently survive, multiply and spread among the haploid nuclei. Thus in some fungi it is possible to have different kinds of haploid nuclei in the same soma and a mixture of haploid and diploid nuclei. In most fungal individuals, the haploid and diploid phases of the life cycle are clearly distinguishable.

Lect.12:

Dr. Warka Saeed Qassim Al-Taee

Sexual spores

The sexual spores are formed as a result of fusion between two opposite sex gametes. They are resting spores, incapable of germination immediately after formation. Sexual spores are oospores, zygospores, ascospores and basidiospores desiring their names from the class to which the fungi belong.

1. Oospores An oospore (Gr.oon = egg + spora = seed, spore) is a sexually produced spore, which develops from unequal gametangial copulation or markedly unequal gametic fusion. It is the characteristic sexually produced spore of oomycetes. Oospores develop from fertilized oospheres (Gr. oon = egg + sphaira = sphere). One or more oospheres develop within 'oogonia', which are multinucleate, globose and female gametangia



2. Zygospores

Zygospores (Gr. zygos = yoke + spora = seed, spore) are sexually produced resting spores or structures formed as a result of plasmogamy between two gametangia, which are usually equal in size. They are

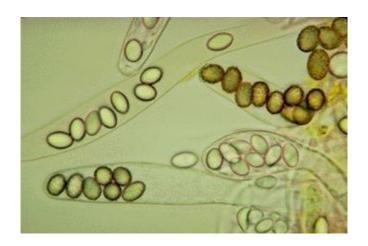
resting structures. Zygospores are the typical sexually produced spores of Zygomycetes e.g. Mucorales and Entomophthorales. Zygospores are often large, thick-walled, warty structures with large food reserves and are unsuitable for long distance dispersal.

3. Ascospores Ascospores

(Gr. askos = sac + spora = seed, spore) are the characteristic spores of the large group of fungi known as Ascomycotina. They are formed as a result of nuclear fusion immediately followed by meiosis. The four haploid daughter nuclei then divide mitotically to give eight haploid nuclei around which the ascospores are cut out. In most ascomycetes, the eight ascospores are contained within a cylindrical sac or ascus from which they are forcible ejected by a squirting process in which the ascus contents, consisting of ascospores and ascus sap, are ejected by explosive breakdown of the tip of the turgid ascus whose elastic walls contract.

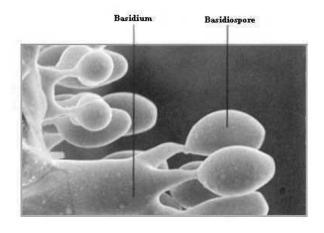
Ascus

Ascus (pl. asci; Gr. askos = sac) is a sac-like cell generally containing a definite number of ascospores (typically eight) formed by free cell formation usually after karyogamy and meiosis. In the large majority of the Ascomycetes the asci are elongated, either club-shaped or cylindrical. But globose, ovoid or rectangular asci are also found. Ordinarily the ascus represents a single cell in which the ascospores are formed. Asci may be stalked or sessile, they may arise from a common fascicle and spread out like a fan or they may arise simply at various levels within the fruiting body.



4. Basidiospores

Basidiospore (Gr. Basidion = small base + spora = seed, spore) is a spore borne on the outside of a basidium, following karyogamy and meiosis. Basidiospores are more uniform compared to ascospores. Typically they are unicellular, but transversely septate spores are found in certain groups like Dacrymycetaceae (Reid, 1974) In shape, they vary from globose, sausage-shaped, fusoid, almond-shaped (i.e. flattened) and the wall may be smooth or ornamented with spores, ridges or folds. The colour of basidiospores is an important criterion of classification.



Asexual fructifications

In fungi conidiophores are grouped together to form specialized structures such as synnemata (sing. synnema) and sporodochia (sing. sporodochium) or produced in fructifications known as pycnidia (sing. pycnidium) and acervuli (sing. acervulus).

a. Synnema or coremium

Synnema or Coremium (pl. coremia) Consists of a group of conidiophores often united at the base and part way up the top. Conidia may be formed along the length of the synnema or only at its apex. The conidiophores comprising a synnema are often branched at the top with the conidia arising from the conidiogenous cells at the tips of the numerous branches. e.g. Deuteromycotina (Arthrobotryum sp (Fig), Penicillium claviforme, Doratomyces stemonitis, Ceratocystis ulmi.





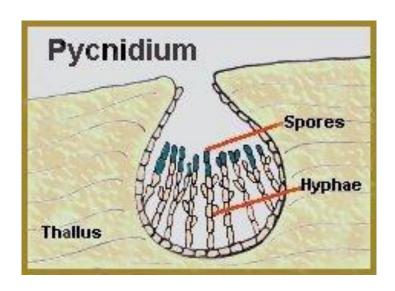
b. Sporodochium

Sporodochium is a fruiting body in which conidiophores arise from a central cushion-like aggregation of hyphae. The conidiophores are packed tightly together and are generally shorter than those composing a synnema. e.g. Epicoccum, Nectria. sporodochium: (pl. sporodochia)

Superficial, cushion-shaped asexual fruiting body consisting of a cluster of conidiophores.

c. Pycnidium

Pycnidium is a globose or flask-shaped body, which is lined on the inside with conidiophores. e.g. Septoria ,Phoma, Ascochyta, Leptosphaeria. Pycnidia may be completely closed or may have an opening. The opening or mouth of pycnidium is called ostiole (L. ostiolum = little door). They may be provided with a small papilla or with a long neck leading to the opening. Pycnidia vary greatly in size, shape, colour and consistency of the pseudoparenchymous wall. The wall of pycnidium is called peridium (pl. peridia; G. peridion=small leather pouch) and it is composed of multicellular layer, as fungal tissues. Pycnidia may formed superficially or sunken in the substratum. They may be formed directly by the loose mycelium or may be definitely stromatic.



d. Acervulus

Acervulus (pl. acervuli) is a fruiting structure commonly found in the order Melanconiales (Deuteromycotina). It is typically a flat or saucershaped mass of aggregated hyphae bearing short conidiophores in a compact layer. Intermingled with the conidiophores, setae (sing. seta; L. seta = bristle) are found. Setae are long, pointed, dark coloured, sterile structures. In nature acervuli are produced on plant tissues subepidermally or subcuticularly and becomes erumpent on maturity. e.g. Colletotrichum.



Sorus

Sorus (pl. sori; Gr. Soros = heap) is a little heap of sporangia or spores. It may be naked or covered by a thin false membrane, as in smuts, or protected by the epidermis as in rust diseases or white blister or white rust (Albugo spp.). The structures break open at maturity and release the spores within, in the form of rust, which is characteristic of these diseases.

Biochemical targets for antifungal agents:

- •Antifungal chemotherapy depends on biochemical differences between fungi and mammal cells. Such differences are few since both type of cells are eukarytoics. Example of such differences
- 1.Fungal cells have both cell membrane and outer cell wall, where as mammalian cells only have cell membrane
- 2.The membranes of fungal and mammalian cells have different types of sterols (i.e. Ergosterol and cholesterol, respectively).

β-glucan component of the fungal cell wall

The structural carbohydrate polymers glucan and chitin compliment and reinforce each other in a dynamic process to maintain the integrity and physical strength of the fungal cell wall. The assembly of chitin and glucan in the cell wall of the budding yeast Saccharomyces cerevisiae and the polymorphic human pathogen Candida albicans are essential processes that involve a range of fungal-specific enzymes and regulatory networks. The fungal cell wall is, therefore, an attractive target for novel therapies as host cells lack many cell wall-related proteins. The most recent class of antifungal drug approved for clinical use, the echinocandins, targets the synthesis of cell wall $\beta(1-3)$ glucan. The echinocandins are effective at treating invasive and bloodstream Candida infections and are now widely used in the clinic. However, there have been sporadic reports of breakthrough infections in patients undergoing echinocandin therapy. The acquisition of point mutations in the FKS genes that encode the catalytic $\beta(1-3)$ glucan synthase subunits.

Ergosterol:

Is sterol found in cell membranes of fungi and protozoa, serving many of the cell functions that cholesterol seres in animal cells.

Because many fungi and protozoa cannot survive without ergosterol, the enzymes that create it have become important targets for drug discovery.

Biochemical targets for antifungal agents (Ergosterol):

Cholesterol

Ergosterol

•Although the ergosterol and cholesterol are quite similar, the side chains are slightly different, and when three-dimensional models are constructed, the ring system of ergosterolis slightly flatter because of the additional double bonds in the B ring.

3

- •This difference in sterol components provides the biochemical basis of selective toxicity for most of the currently available antifungal drugs.
- The antifungal agents can be divided into the

following classes, based on their chemical structure, mechanism of action, and source:

I.Antibiotics: Amphotericin B, Nystatin, Griseofulvin

II.Azoles(imidazole, triazole derivates)

- •Imidazoles—Clotrimazole, Ketoconazole, Miconazole, Bifonazole, Butoconazole, and Zinoconazole
- •Triazoles—Fluconazole, Itraconzole, Terconazole

III.AllylaminesTolnaftate, Naftifine, and Terbinafine

IV.Fluorinated pyrimidines: Flucytosine

V.Chitin synthetase inhibitors: NikomycinZ

VI.Peptides/proteins: Cispentacin

VII.Fatty and other acids: propionic acid,

undecylenic acid,

Polyene membrane disrupters:

- •The polyenes have an affinity to sterolcontaining membranes, thus being inserted into the membrane, causes leakage and disruption of function.
- •The polyenes have higher affinity for ergosterol over cholesterol-containing membrane.
- •NYSTATIN Nystatin, the first clinically useful polyene antifungal antibiotic, is a conjugated tetraene isolated from cultures of the bacterium *Streptomyces noursei S. nodosus* in 1951. It is too toxic for systematic use and can be used orally to treat yeast infections.
- •AMPHOTERICIN B Amphotericin B, which as a heptaene has low enough toxicity to mammalian cells to permit intravenous (IV) administration, was discovered in 1956. It can not cross bloodbrain barrier. Formulated as water-soluble complex with deoxycholic acid for IV administration.
- Azole antifungal are the largest class of antimycotics available today.

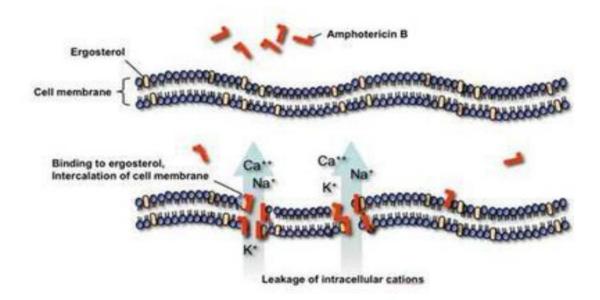
- •Some azoles are used topically, for v dermatophytic infections others are used orally to treat systematic infections.
- •Unlike amphotericin B, azoles are orally bio available and have broader spectrum of activity.
- •All azoles inhibit 14α -demethylase of ergosterol biosynthesis
- •At high in vitro concentrations (micromolar), the azoles are fungicidal; at low in vitro concentrations (nanomolar), they are fungistatic.

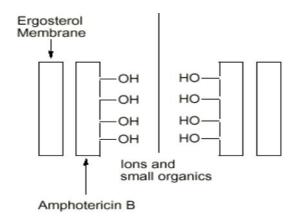
Ergosterol Biosynthesis Inhibitors:

- •The function of lanosterol 14α -demethylase is to oxidatively remove a methyl group from lanosterol during ergosterol biosynthesis.
- •The enzyme is membrane-bound of the class cytochrome P450.
- •The enzyme possesses a heme moiety as part of its structure, and the basic electron pairs of the azole rings can occupy a binding site and prevent the enzyme from turning over
- •The enzyme is also present in mammalian

biosynthesis of cholesterol, and the azoles are known to inhibit cholesterol biosynthesis also (e.g. biosynthesis of adernocorticoids)

- •The mammalian copy of the enzyme is much sensitive and binds azoles with lower affinity than fungal copy (which explains the selective fungal toxicity).
- •The 1,2,4-triazoles appear to cause a lower incidence of endocrine effects and hepatotoxicity than the corresponding imidazoles, possibly because of even lower affinity to mammalian copy of the enzyme.





Mycotoxins Definition

Mycotoxins are toxic secondary metabolic products of molds present on almost all agricultural commodities world wide. Unlike primary metabolites (sugars, amino acids and other substances), secondary metabolites are not essential in the normal metabolic function of the fungus. Other known secondary metabolites are phytotoxins and antibiotics.

Currently there are around 400 mycotoxins reported. These compounds occur under natural conditions in feed as well as in food. Some of the most common mycotoxins include: aflatoxins, trichothecenes, fumonisins, zearalenone, ochratoxin and ergot alkaloids. Mycotoxins are produced by different strains of fungi and each strain can produce more than one mycotoxin.

Each plant can be affected by more than one fungus and each fungus can produce more than one mycotoxin. Consequently, there is a high probability that many mycotoxins are present in one feed ingredient, thus increasing the chances of interaction between mycotoxins and the occurrence of synergistic effects, which are of great concern in livestock health and productivity. Synergistic effects occur when the combined effects of two mycotoxins (even at low levels) are greater than the individual effects of each toxin alone. Simple additive effects can also occur with the combined effects of two mycotoxins being equal to the sum of the effects of each toxin on its own.

Mycotoxins are invisible, tasteless, chemically stable and resistant to temperature and storage. They are resistant the normal feed manufacturing processes.

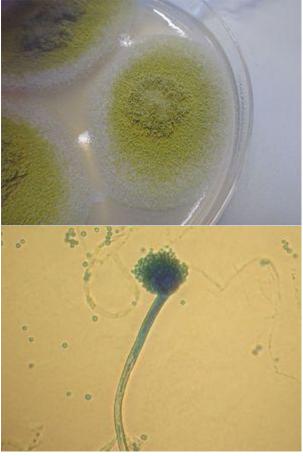
Mycotoxin producing fungi can be divided into two groups.

- Field fungi (such as *Fusarium sp.*) typically produce mycotoxins in the field ("pre-harvest")
- Storage fungi (such as *Aspergillus* and *Penicillium sp.*) typically occur after harvest ("post-harvest")

However, in special cases like under unusually hot or dry conditions *Aspergillus* and *Penicillium* species can also affect crops during the growing season. On the other hand, field fungi can continue growing and produce mycotoxins during transport and storage.

Mycotoxins cause economic losses at all levels of food and feed production, including crop and animal production, processing and distribution. According to the FAO (Food and Agriculture Organization) around 50% of the world's crop harvests may be contaminated with mycotoxins.

Major classes of mycotoxin producing fungi and mycotoxins



A. flavus A. parasiticus

A. nomius

Aflatoxin (B_1, B_2, G_1, G_2)

Α.

pseudotamarii

A. ochraceus Ochratoxin (Ochratoxin A)

A. clavatus

A. ctavatus
A. terreus
Patulin

A. flavus A. versicolor Cyclopiazonic acid (CPA)

Claviceps spp.



C. purpurea C. fusiformis C. paspali C. africana alkaloids:
Clavines
(Argroclavine)
Lysergic acids
Lysergic acid
amides
(Ergine)
Ergopeptines

Ergot

Major classes of mycotoxin producing fungi and mycotoxins

(Ergotamine, Ergovaline)

Fusarium spp.



F. verticillioides (syn. F. moniliforme) F. proliferatum

Fusaric acid

Type A

F. graminearum

Trichothecenes:

F. avenaceum F. culmorum

T-2 toxin, HT-2 toxin, diacetoxyscirpenol

Fumonisin (B1, B2, B3)

F. poae Type B

F. equiseti

Trichothecenes:

F. crookwellense

F. acuminatum
F. sambucinum

Nivalenol, Deoxynivalenol,

F. sporotrichioides Fusarenon-X

F. graminearum

F. culmorum

Zearalenone

F.sporotrichioides

Penicillum spp.

P. verrucosum Ochratoxin
P. viridicatum (Ochratoxin A)

P. citrinum
P. verrucosum

Citrinin

P. roqueforti

Roquefortine C PR toxin

Penitrem A

Major classes of mycotoxin producing fungi and mycotoxins



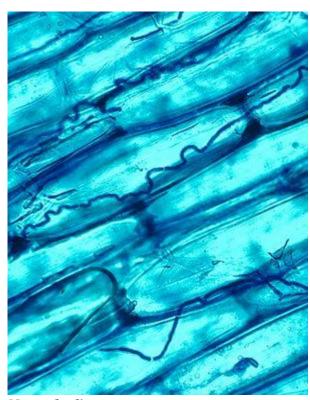
P. cyclopium P. camemberti Cyclopiazonic acid (CPA) Penitrem A

P. expansum

P. claviforme

P. roquefortii

Patulin



Neotyphodium

Tall fescue toxins: N. coenophialum

Ergot alkaloids, Lolines, Peramine

Ryegrass toxins:

N. lolii

Lolitrems, Peramine, Ergot alkaloids (e.g. Ergovaline)



P. chartarum

Pithomyces

Primary mechanism of action Mycotoxin

Binds to guanine (DNA-adduct) after Aflatoxin

metabolic activation in the liver

TrichothecenesInhibition of protein synthesis

Binds to mammalian estrogen receptor Zearalenone

Ochratoxins Blocks protein synthesis

Ergot alkaloids Binding to adrenergic, dopaminergic and

serotonin receptors

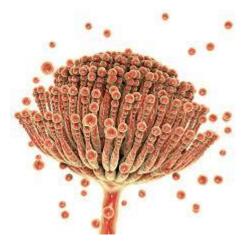
Inhibit ceramide synthase (sphingolipid **Fumonisins**

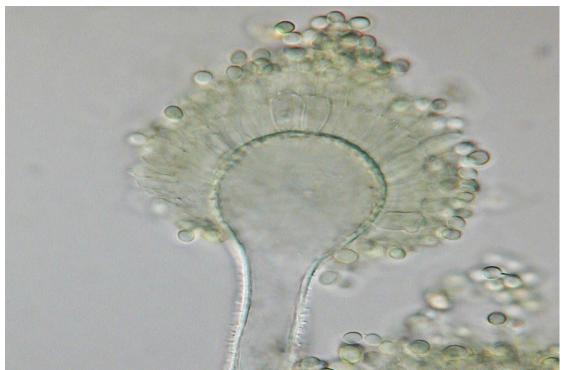
biosynthesis)

Mycotoxins are capable of direct-target toxicity towards certain organs such as the liver, nervous system, kidney, skin, cardiovascular, reproductive and immune systems. Non-direct-target effects include carcinogenicity, teratogenicity and mutagenesis.

Aflatoxins:

Aflatoxins are a type of mycotoxin produced by Aspergillus species of fungi, such as A. flavus and A. parasiticus. The umbrella term aflatoxin refers to four different types of mycotoxins produced, which are B₁, B_2 , G_1 , and G_2 . Aflatoxin B_1 , the most toxic, is a potent carcinogen and has been directly correlated to adverse health effects, such as <u>liver cancer</u>, in many animal species. Aflatoxins are largely associated with commodities produced in the tropics and subtropics, such as cotton, peanuts, spices, pistachios, and maize.





ingdom: Fungi

Division: <u>Ascomycota</u>

Class: <u>Eurotiomycetes</u>

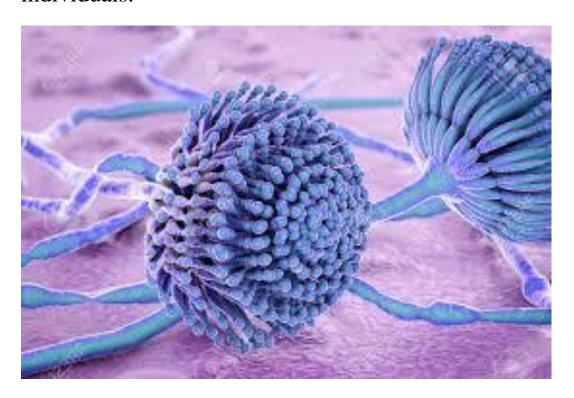
Order: <u>Eurotiales</u>

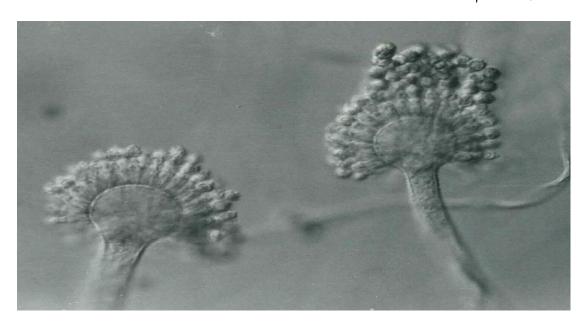
Family: <u>Trichocomaceae</u>

Genus: Aspergillus

Species: A. flavus

Aspergillus flavus is a saprotrophic and pathogenic fungus with a cosmopolitan distribution. It is best known for its colonization of cereal grains, legumes, and tree nuts. Postharvest rot typically develops during harvest, storage, and/or transit. A. flavus infections can occur while hosts are still in the field (preharvest), but often show no symptoms (dormancy) until postharvest storage and/or transport. In addition to causing preharvest and postharvest infections, many strains produce significant quantities of toxic compounds known as mycotoxins, which, when consumed, are toxic to mammals. A. flavus is also an opportunistic human and animal pathogen, causing aspergillosis in immunocompromised individuals.





Kingdom: Fungi

Division: Ascomycota

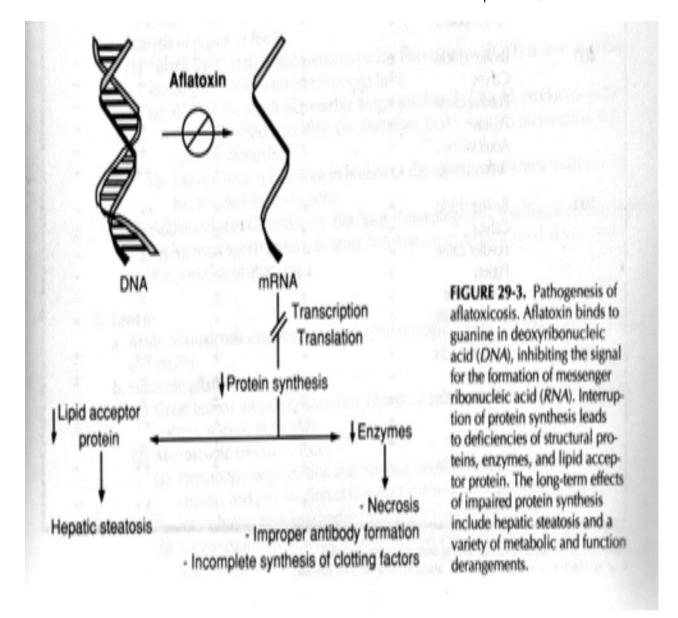
Class: Eurotiomycetes

Order: Eurotiales

Family: <u>Trichocomaceae</u>

Genus: <u>Aspergillus</u>
Species: A. parasiticus

Discovery of mycotoxins \square Serious worldwide concern began in the early 1960s after "Turkey X disease" was discovered in UK. \square More than 100,000 young turkeys on poultry farms died in the course of a few months. \square Investigation of the early outbreaks showed that they were all associated with feeds, namely Brazilian peanut meal .



Ochratoxin:

Ochratoxin is a mycotoxin that comes in three secondary metabolite forms, A, B, and C. All are produced by *Penicillium* and *Aspergillus* species. The three forms differ in that Ochratoxin B (OTB) is a nonchlorinated form of Ochratoxin A (OTA) and that Ochratoxin C (OTC) is an ethyl ester form Ochratoxin A. The largest amounts ochratoxins are

made by A. ochraceus and P. cyclopium.

A. ochraceus and P. viridicatum (reclassified as

P. verrucosum), two species that were first reported ochratoxin A (OA) producers, occur most frequently in nature. Other fungi, such A. citricus, and A. Petromyces alliceus, fonsecaeus (both in A. niger group), have also been found to produce OA. Most of the OA producers are storage fungi and preharvest fungal infection. Aspergillus ochraceus is found as a contaminant of a wide range of commodities including beverages such as beer and wine. Aspergillus carbonarius is the main species found on vine fruit, which releases its toxin during the juice making process. OTA has been labeled as a carcinogen and a nephrotoxin, and has been linked to tumors in the human urinary tract, although research in humans is limited confounding factors.

Although most OA producers can grow in a range from 48C to 37C and at aw as low as 0.78, optimal conditions for toxin production are narrower with temperature at 24–25C and aw values .0.97.

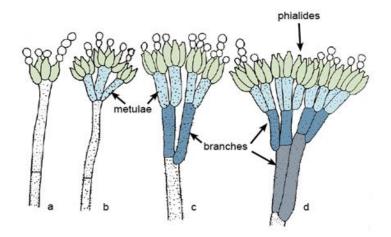
Ochratoxins are produced primarily in cereal grains (barley, oats, corn, wheat) and mixed feed during storage in temperate climatic conditions, with levels higher than 1 ppm being reported.

OA has been found in other commodities, including beans, coffee, nuts, olives, raisin, cheese, fish, pork, milk powder, fruit juices wine beer, peppers.

OA can be carried through the food chain because of the presence of OA residues in animal products as result of its binding with serum albumin. Natural occurrence of OA in kidneys, blood serum, blood sausage.

Citrinin:

Citrinin is a toxin that was first isolated from *Penicillium citrinum*, but has been identified in over a dozen species of *Penicillium* and several species of *Aspergillus*. Some of these species are used to produce human foodstuffs such as cheese (*Penicillium camemberti*), sake, miso, and soy sauce (*Aspergillus oryzae*). Citrinin is associated with yellowed rice disease in Japan and acts as a nephrotoxin in all animal species tested. Although it is associated with many human foods (wheat, rice, corn, barley, oats, rye, and food colored with Monascus pigment) its full significance for human health is unknown. Citrinin can also act synergistically with Ochratoxin A to depress RNA synthesis in murine kidneys.



Ergot Alkaloids:

1. Source: Claviceps purpurea /barley, wheat & oats

2. Factor favoring: Warm & humid

Mechanisim of toxic:

a. potent initiators of contraction in smooth muscle

b. mimic the action of dopamine.

Clinical sign

a. necrosis of the feet, ears and tail

b. increased temperature., pulse & respiration rate

c. lactation does not occur

d. hyper-excitability & tremors

e. heat intolerance in cattle

are compounds produced as a toxic mixture of alkaloids in the <u>sclerotia</u> of species of *Claviceps*, which are common pathogens of various grass species.

The ingestion of ergot sclerotia from infected cereals, commonly in the form of bread produced from contaminated flour, cause ergotism, the human disease historically known as St. Anthony's Fire. There are two forms of ergotism: gangrenous, affecting blood supply to extremities, and convulsive, affecting the central nervous system. Modern methods of grain cleaning have significantly reduced ergotism as a human disease; however, it is still an important veterinary problem. Ergot alkaloids have been used pharmaceutically.

Patulin:

Patulin is a toxin produced by the *P. expansum*, *Aspergillus*, *Penicillium*, and *Paecilomyces* fungal species. *P. expansum* is especially associated with a range of moldy <u>fruits</u> and <u>vegetables</u>, in particular rotting apples and figs. It is destroyed by the <u>fermentation</u> process and so is not found in apple beverages, such as <u>cider</u>. Although patulin has not been shown to be carcinogenic, it has been reported to damage the <u>immune system</u> in animals. In 2004, the <u>European Community</u> set limits to the concentrations of patulin in food products. They currently stand at 50 μg/kg in all fruit juice concentrations, at 25 μg/kg in solid apple products used for direct consumption, and at 10 μg/kg for children's apple products, including apple juice.

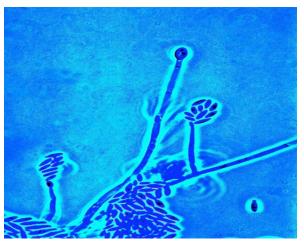
Fumonisins (Fm):

Fumonisins are a group of toxic metabolites produced primarily by *F. verticillioides*, *F. proliferatum* and other related species readily colonize corn all over the world.

Although *F. anthophilum*, *F. nupiforme*, and *F. nygam*ai are capable of producing Fms . More than 11 structurally related Fms (B1, B2, B3, B4, C1, C4, A1, A2, etc.), have been found since the discovery of FmB1.

Fumonisins are most frequently found in corn, cornbased foods, and other grains (such as sorghum and rice). The level of contamination varies considerably with different regions and year, ranging from negligible to more than 100 ppm; but is generally reported to be between 1 and 2 ppm.

Fusarium toxins are produced by over 50 species of *Fusarium* and have a history of infecting the grain of developing cereals such as wheat and maize. They include a range of mycotoxins, such as: the fumonisins, which affect the nervous systems of horses and may cause cancer in rodents; the trichothecenes, which are most strongly associated with chronic and fatal toxic effects in animals and humans; and zearalenone, which is not correlated to any fatal toxic effects in animals or humans. Some of the other major types of *Fusarium* toxins include: beauvercin and enniatins, butenolide, equisetin, and fusarins.

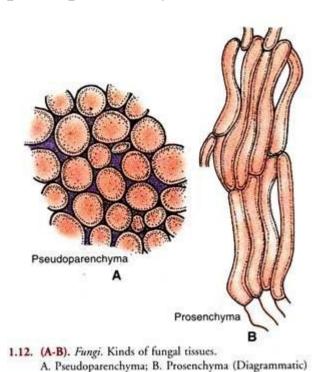




Hyphal modification in fungi

1-prosenchyma:

Is formed by the loosely packed tissue like organization of fungi .is formed when the component hyphae is arranged more or less parallel to one another and whole mass become afelt like structure the individuality of fungal hyphae is not lost . is also known plectenchyma or proso-plectenchyma .



2- Pseudo-parenchyma:

Here hyphae are closely inter twined and not distinguishable from each other . it resembles with the parenchymatous tissue of higher plantes in cross section .

called as para-plectenchyma.

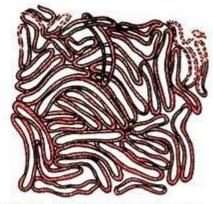


Fig. 1.13. Fungi. Prosenchymatous tissue in the pith of Claviceps purpurea; the hyphal elements are recognisable (Diagrammatic).

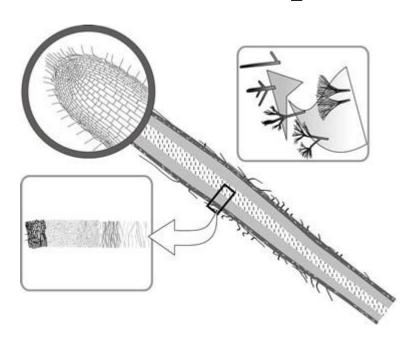
3-Rhizomorphs: (mycelial

cords)

Are thick strands or root like aggregation of somatic hyphae in some fungi called as mycelial cords . they are gelatinous , dark brown and coiled structures . hyphae loose its individuality . hyphae are arranged in parallel way and higher infection capacity than individual hyphae .(high penetration capacity and survives for many years and they give new mycelia in the favourable conditions .



2



4- Sclerotium: (sclerotia).

Is a compact globose structure formed by the aggregation of hyphae in some fungi . the individuality of hyphae is lost and the mass become rounded and cushion like structures and survives for long periods , some times for many years . (resting stage of some fungi)

[outer portion dark brown, inner cells are colour less]



(Ergot in wheat)

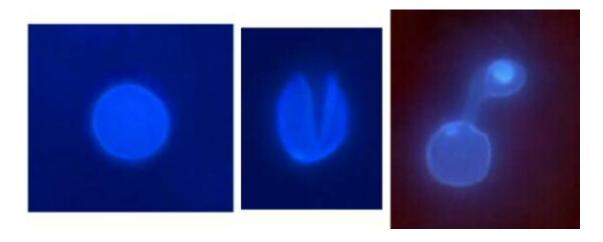
Ergot, fungal disease of cereal grasses, especially rye than wheat, caused by

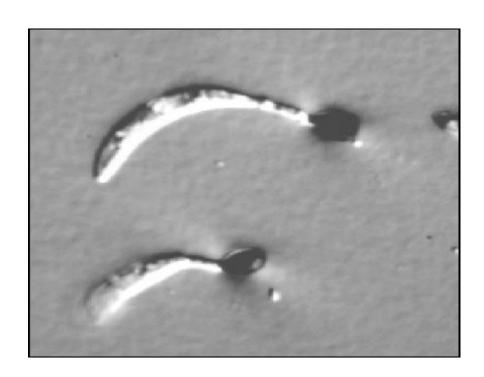
species of the ascomycete fungus Claviceps. The disease decreases the production of viable grains by infected plants and can contaminate harvests. Ergot is commonly associated with rye infected by *C. purpurea*.

3

5-Appressorium:

Is a terminal simple or lobed swollen structure of germ tubes on infecting hyphae. in many parasitic fungi . it adheres to the surface of host and help in the penetrating of hyphae .





4

6-Stroma:

- 1 -Stroma are compact somatic structures.
- 2-They are flat cushion like pseudoparenchymatous structures.
- 3-Fructification are usually found on or in them.



7-Snares (hyphal traps) :

Snares are trap like structures produced by som fungi to capture small animals such nematodes and protozoa .

8-Haustoria.

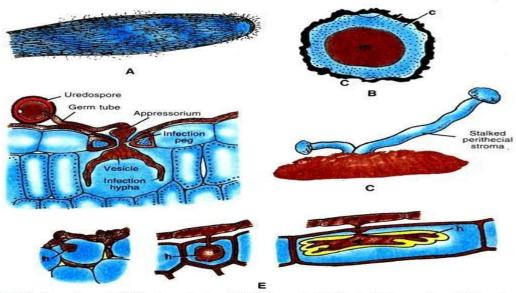


Fig. 1.14. Aggregation and Modifications of hyphae. A-B, Rhizomorph; C, Sclerotia; D, Appressorium; E, Haustorium.