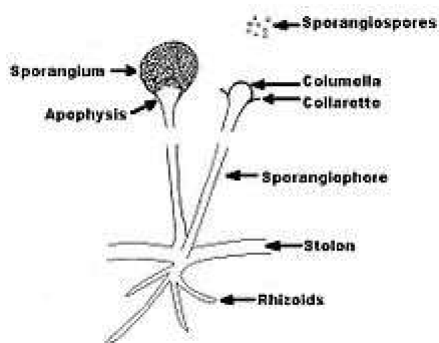


Rhizopus

Rhizopus



Schematic diagram of *Rhizopus* spp.

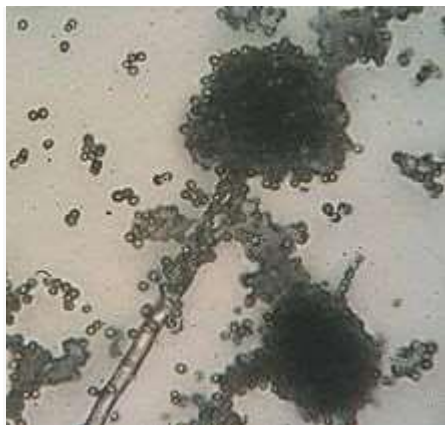
Scientific classification

Kingdom:	Fungi
Division:	Zygomycota
Class:	Mucormycotina
Order:	Mucorales
Family:	Mucoraceae
Genus:	<i>Rhizopus</i> Ehrenb. (1820)

Type species

<i>Rhizopus</i>	<i>nigricans</i>
Ehrenb. (1820)	

Rhizopus is a genus of common saprophytic fungi on plants and specialized parasites on animals. They are found on a wide variety of organic substrates, including "mature fruits and vegetables", jellies, syrups, leather, bread, peanuts, and tobacco. Some *Rhizopus* species are opportunistic agents of human zygomycosis (fungal infection) and can be fatal. *Rhizopus* infections may also be a complication of diabetic ketoacidosis. This widespread genus includes at least eight species.



Rhizopus 400x magnification

Rhizopus Stolonifer: Vegetative Structure and Reproduction

Vegetative Structure of Rhizopus Stolonifer:

The vegetative plant body is eucarpic and consists of white cottony, much branched mycelium. The mycelial plant body is differentiated into nodes and internodes (Fig. 4.25A).

The internodal region is the aerial and arching hyphae, known as stolon, which when touches the substratum forms the nodal region. The nodal region bears much branched rhizoid grows downward, inside the substratum for anchorage and absorption of food.

The hyphal wall is microfibrillar and consists mainly of chitin-chitosan. In addition to chitin-chitosan, other substances like proteins, lipids, purines and salts like calcium and magnesium are also present in the hyphal wall.

Inner to the cell wall, cell membrane is present which covers the protoplast (Fig. 4.25B, C). The protoplast contains many nuclei, mitochondria, endoplasmic reticulum, ribosome, oil droplets, vacuoles and other substances. The size of the vacuole enlarges with age by coalescence of smaller vacuoles.

Reproduction in Rhizopus Stolonifer:

Rhizopus Stolonifer reproduces by vegetative, asexual and sexual means.

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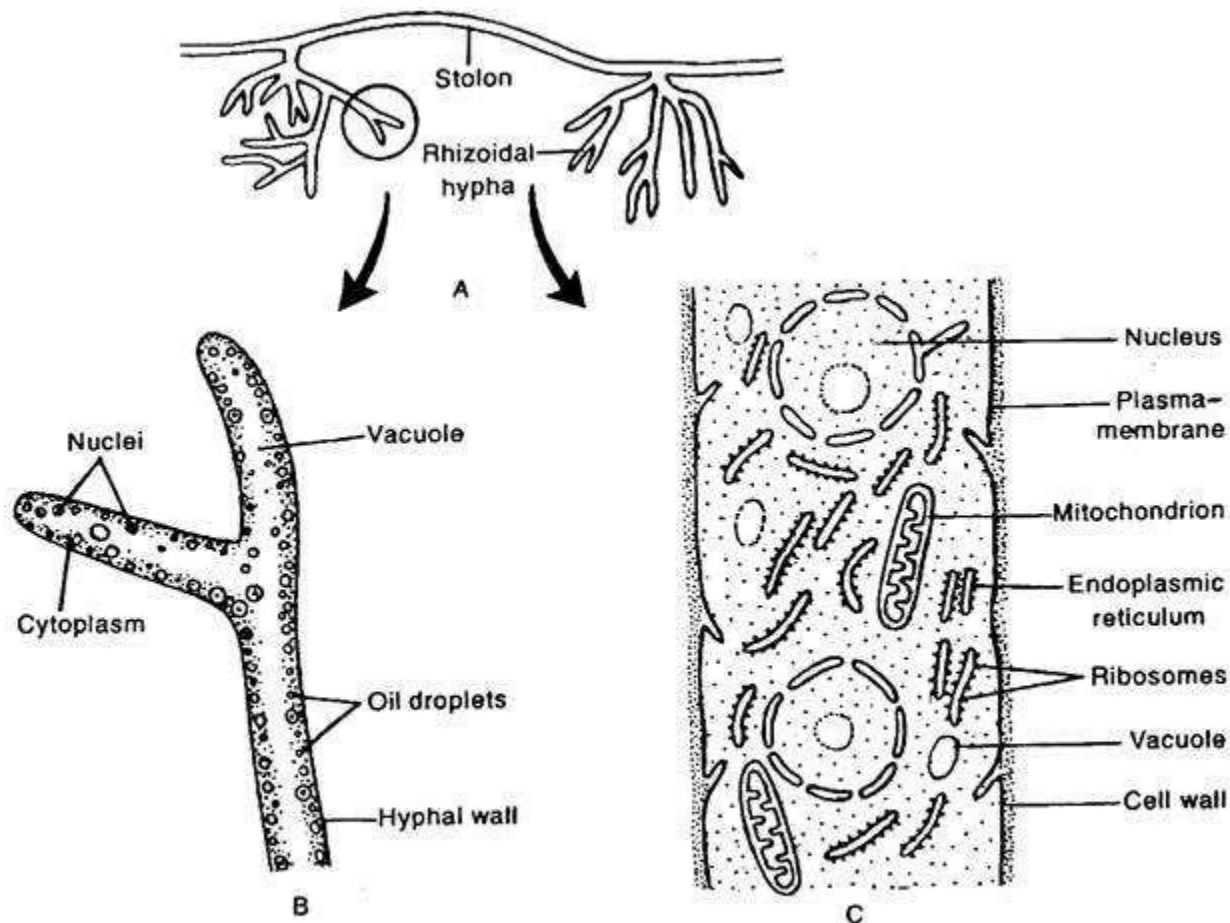


Fig. 4.25 : *Rhizopus stolonifer* : A. Vegetative mycelium, B. Portion of hypha under light microscope, C. Portion of hypha under electron microscope

2. Asexual Reproduction:

It takes place by the formation of sporangiospore and chlamydospore.

(a) Sporangiospore Formation:

During favourable condition, the non-motile spores such as sporangiospores or aplanospores are formed inside the sporangium. The sporangium develops singly at the apex of sporangiophore. The sporangiophore develops in tuft from the upper side of node opposite to the rhizoidal hyphae (Fig. 4.26B). Initially, a number of elongated hyphae develop aerially from the upper- side of the node which elongate upto a certain height.

The nuclei and cytoplasm push more and more towards the apical side, consequently the apex of the aerial hyphae swells up (Fig. 4.26C). The swollen part enlarges and develops into a large round sporangium (Fig. 4.26D)

It takes place by fragmentation. Due to accidental breakage the stolon may break up into two or more small units. Each unit is capable of growing as mother mycelium.

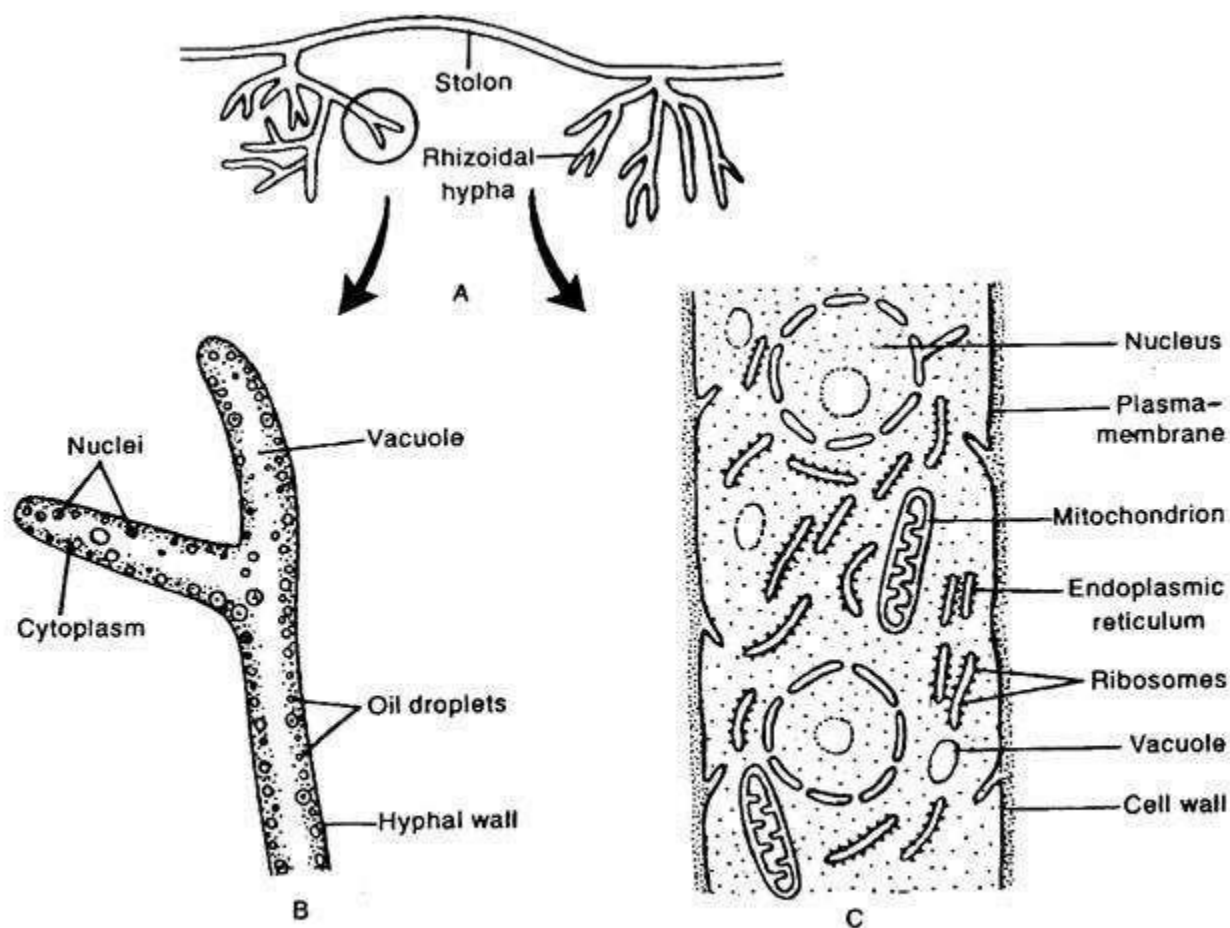


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Venturia inaequalis

Venturia inaequalis is an ascomycete fungus that causes the Apple scab diseases. *Venturia inaequalis* anamorphs have been described under the names *Fusicladium dendriticum* and *Spilocaea pomi*. Whether *V. inaequalis* is a single species or contains several cryptic species has been a matter of debate for a long time. Recent genetic studies have revealed a considerable uniformity of the species. In addition, the fungus *Spilocaea pyracanthae*, a parasite of *Pyracantha* appeared not to genetically differ from *V. inaequalis*, being thus a special form of the latter.

Morphology

The fruiting bodies, ascocarps appear in the form of pseudothecia. They are solitary and embedded into the host plant tissue. A pseudothecium has small dark hairs around its opening, and contains pseudoparaphyses along with asci. The asci contain eight haploid ascospores. The haploid chromosome number of *V. inaequalis* is seven.

Life cycle

The infection cycle begins in the springtime, when suitable temperatures and moisture promote the release of *V. inaequalis* ascospores. These spores rise into the air and land on the surface of a susceptible tree, where they germinate and form a germ tube that can directly penetrate the plant's waxy cuticle. A fungal mycelium forms between the cuticle and underlying epidermal tissue, developing asexually the conidia, that germinate on fresh areas of the host tree, which in turn produce another generation of conidial spores. This cycle of secondary infections continues throughout the summer, until the leaves and fruit fall from the tree at the onset of winter.

V. inaequalis overwinters mostly as immature Perithecia, where sexual reproduction takes place, producing a new generation of ascospores that are released the following spring. Scab lesions

located on the woody tissues may also overwinter in place, but will not undergo a sexual reproduction cycle; these lesions can still produce ineffective conidial spores in the spring.

Apple scab occurs everywhere in the world where apples are grown and Causes more losses than any other apple disease. It is most serious in areas that have cool, wet weather during the spring and may not be economically important in warm or dry climates.

Symptoms and Signs

Apple scab results in symptoms on the aerial parts of the apple tree, including Leaves, petioles, flowers, sepals, fruit, pedicels, young shoots, and bud scales

Apple leaves and fruit

The symptoms are generally most noticeable and serious on leaves and fruit. The first lesions seen in the spring are usually on the underside of expanding leaves. Once the leaves open, the upper surfaces also become vulnerable to infection. A lesion first appears as an area which is a lighter shade of green than the surrounding leaf. The lesion is usually circular and as it increases in size it becomes olive colored and velvety due to production of asexual spores (conidia). Lesions that form on young leaves may be quite large, some more than 1 cm in diameter. Lesions that form on expanded leaves are usually smaller because older leaves are more resistant to infection. Affected tissues eventually may become distorted and puckered, and the leaf lesions often become cracked and torn. Lesions on the leaves and fruit are generally blistered and "scabby" in appearance, with a distinct margin. The earliest noticeable symptom on fruit is water-soaked areas which develop into velvety, green to olive brown lesions. Infections of young fruit will cause fruit distortion. Severely infected leaves or fruit will often drop from the tree. Infection which causes significant defoliation for two or three years in a row can result in weakened trees that are more susceptible to freeze damage, insect injury, and other diseases.

Apple blossoms

Symptoms on blossoms usually occur as small, dark green lesions at the base of the flower, on the sepals, and on the stem pedicel before and during bloom. When pedicels become infected the developing fruit may drop, resulting in lower fruit yield.

Pathogen Biology

Venturia inaequalis is an ascomycete fungus because it produces sexual spores (ascospores) in a sac-like structure called an ascus (plural asci). The mycelium of *V. inaequalis* is septate, and the nuclei are haploid.

Sexual Reproduction

Venturia inaequalis occurs as two mating types. Both must be present in order for sexual reproduction to be initiated. Mating takes place in the early spring within infected fallen leaves left from the previous season. It consists of fusion of a male organ (antheridium) formed from a hyphal tip of one mate to a female receptive hypha (trichogyne) from the opposite mate. The trichogyne is attached to a coil of hyphae called the pseudothecial initial. During fertilization, a nucleus passes from the antheridium through the trichogyne into a cell at the base of the pseudothecial initial. After fertilization, the pseudothecial initial develops into a pseudothecium. The pseudothecium is a cavity located within a dense mat of fungal mycelium called a stroma. Inside this cavity the asci and ascospores are formed. The very brief diploid stage in the life cycle of *V. inaequalis* occurs within the pseudothecium in single hyphal cells (croziers) which give rise to the haploid ascospores following meiosis. The asci are elongated, sac-like structures, each of which contains eight ascospores in a linear arrangement. The ascospores are brown, two-celled, and have a characteristic "footprint" shape. The shape of the ascospores inspired the Latin name for apple scab, "inaequalis", which refers to the unequal size of the cells. They measure between 5 and 7 μm wide and between 11 and 15 μm long. In the spring, when the fallen leaves become wet, the mature pseudothecia swell and protrude from the surface of the leaf. The ascospores are released and carried by rain and wind. If they land on the young blossoms and leaves of apple trees, they germinate and initiate infection. There is only one cycle of ascospore production and infection within a season.

Asexual Reproduction

Like most other ascomycetes, *V. inaequalis* reproduces asexually by producing spores called conidia. The conidial stage of the *V. inaequalis* lifecycle has its own name, *Spilocaea pomi*. The conidia are single-celled, uninucleate, brown or olive in mass but lighter when viewed individually under a microscope, and are narrower at one end than the other. They measure between 6 and 12 μm wide and between 12 and 22 μm long. Conidia are produced by specialized short hyphae called conidiophores. The conidiophores are formed on a dense mat of mycelium which pushes up through the leaf cuticle and ruptures it. It is the mass of conidia and conidiophores which causes the velvety appearance of the young lesions. Conidia are

produced within the lesions from nine to thirty days after initial infection of the leaf, depending on the temperature. Conidia are disseminated by wind and by windblown and splashing rain. Once a conidium lands on an apple leaf, blossom or fruit, it adheres to the surface and germinates, provided the tissue is wet. The germination hypha penetrates the cuticle and establishes a new infection. There can be many cycles of conidial production and infection within a single growing season. When infection of fruit occurs late in the fall it may not be detected until the fruit has been stored for several months.

Disease Cycle

In living leaves, the growth of the apple scab fungus is confined to the area between the host cuticle and the epidermis. It is not clear where the fungus obtains nutrients since it does not grow into the leaf cells, but it is generally believed that it degrades and utilizes the cuticle itself as well as some host cell wall and pectic material. Once infected leaves have fallen and died, the fungal mycelium colonizes them completely. *Venturia inaequalis* usually overwinters in the fallen leaves as mycelium and pseudothecial initials. In milder climates, the fungus may overwinter in infected buds and produce conidia which serve as primary inoculum. In leaf debris, most of the pseudothecial initials form within a month after the leaves fall, and then enter a period of dormancy. In the spring, mating takes place in the leaves between two mating types. When the mature pseudothecia become wet, the asci expand through the top of the pseudothecium. The ascospores are forcibly discharged into the air where they are spread by the wind and by windblown rain. If there is sufficient moisture from rain or dew, the ascospores infect and produce lesions on apple blossoms and young unfolding leaves. In most years, ascospore release coincides with the several week period from budbreak through the end of bloom. From 9 to 30 days later, the primary lesions produce conidia, which are dispersed to other leaves and developing fruit where they establish secondary infections. Up to 100,000 conidia can be produced by a single lesion. Lesions from ascospore or conidial infection expand at a rate which is determined partly by temperature and partly by characteristics of the host tissue, including genotype and age. The rate of lesion expansion in turn affects the speed with which new spores are produced. During a cooler season, or on more resistant apple trees, lesions expand more slowly and may be smaller in size. As a result, the number of secondary cycles will be fewer.

The production of conidia is affected by humidity: levels of at least 60-70% are required for spore production. Infection of leaves by ascospores and conidia is highly dependent on how long the

leaves or fruit stay wet, and on the average temperature. The Mills table relating leaf wetness duration and temperature is used to determine the likelihood that infection will occur if conidia are present. For example, at an average temperature of 18° C (65° F), light infection will result if leaves remain wet for nine hours, but if leaves are wet for 18 hours or more heavy infection will occur. Lesions will produce conidia after nine days if the temperature averages 18° C (65° F), but not until 17 days if the temperatures are lower, averaging only 8° C (49° F). The Mills table continues to be revised as more data are gathered from different regions.

Disease Management

Chemical Control

Management of apple scab on susceptible trees is focused on the prevention of primary infection by ascospores in the spring. Early infection of trees may result in poor fruit set, and will result in more secondary inoculum being produced throughout the season. The initial fungicide sprays are therefore timed to coincide with the spring release of primary inoculum. Later sprays are often targeted at other fungal diseases, in addition to scab, but also are effective against apple scab secondary inoculum. In commercial orchards, airblast sprayers are typically used to apply fungicides. Chemical fungicides for apple scab control are generally of two types, preventive (protectant) and curative. Most growers use a combination of the two for maximum effectiveness. Preventive fungicides are applied to the leaves and fruit before infection. When ascospores or conidia are present on susceptible plant surfaces and leaf moisture and temperature are suitable, the fungicide prevents the fungal spores from germinating or penetrating the host tissue. Good spray coverage and uniform deposition are essential. To ensure coverage of newly emerging tissues and to replace the chemical as it is lost to weathering, fungicides must be reapplied on a regular schedule. Usually this consists of spraying every 5 to 7 days in the early part of the season, and every 7 to 10 days later in the season. Curative fungicides have limited systemic activity and if applied soon after infection, are capable of penetrating the leaf and stopping further development of the fungus, thus preventing symptom development. These fungicides are especially useful when growers use a Mills table based weather monitoring system that tells them when an infection has occurred. Prompt application of the fungicide will stop further disease development. Growers using these systems spray fungicides only when needed, and in a season when few infections occur (in a dry spring for example), they may apply fewer

fungicide treatments than a grower spraying preventively. By the time apples are harvested in summer and fall, fruit fungicide residues are either absent or present at extremely low safe levels

Significance

Apple scab disease has long been a problem on apples; symptoms of the disease can be easily identified on fruit in paintings from the fifteenth and sixteenth centuries. The frequent depictions of scabinfected apples suggest that the disease was common and that the affected fruit was acceptable in earlier times. All the commonly grown apple varieties were susceptible to the disease, and there were no chemical treatments until the late 1800s. At that time, copper and Sulfur based fungicides provided preinfection protection, but the treatments caused substantial damage to the apple foliage. Even today, in spite of the highly effective chemicals and the resistant apple varieties that are available, apple scab causes greater economic losses of apples in North and South America, Europe, and Asia than any other apple disease. *Venturia inaequalis* was one of the first ascomycetes to be subjected to genetic analysis; the heritability of pathogenicity and of sexual compatibility was investigated as early as the 1930s. It has served as and continues to be a valuable tool for basic genetic research and for studies of the inheritance of pathogenicity. Among the features that make *Venturia inaequalis* so amenable for genetic research is the fact that it is similar to many obligate parasites which infect young tissues and live in a close association with them, without visibly harming them, for an extended period of time. Yet unlike obligate parasites, it can be cultured on artificial media, and matings can be made *in vitro*. Other advantages of *V. inaequalis* for genetic studies include its considerable diversity, with many pathotypes or races occurring in the natural population; the stability of phenotypes and genotypes in culture over many years; the fact that it is haploid, allowing the effects of alleles to be studied directly; the uninucleate conidia which give rise to colonies in which all the nuclei are genetically uniform; and the ability to isolate all eight of the ascospores from a single ascus. The eight spores are the result of a single meiotic event, and so segregation of traits can be studied directly rather than statistically. Furthermore, the eight ascospores are arranged in order, allowing reconstruction of the meiotic divisions and mapping of centromeres as genetic loci. Many of these advantages are shared by other Ascomycetes, but they are uniquely brought together in *V. inaequalis*. Many years of study have demonstrated that the genetics of pathogenicity in *V. inaequalis* are not simple. There are multiple genes involved in

pathogenicity to various apple species and cultivars. In several of these interactions, pathogenicity is conditioned by single genes. In some cases, there appears to be a gene-for-gene relationship between the host and the pathogen. In a gene-for-gene interaction, each gene for resistance in the host is matched by a gene for pathogenicity (or virulence) in the pathogen. Resistance will only occur if the host has the dominant allele for the resistance gene and the pathogen has the corresponding dominant allele conditioning nonpathogenicity (or avirulence); all other combinations will result in disease. Gene-for-gene interactions are found in numerous host-pathogen interactions, and are thought to be the consequence of co-evolution of the host and the pathogen.

Because the effects of various environmental parameters on the development of apple scab epidemics are well understood and fairly predictable, this disease serves as a useful model for the teaching of important epidemiological principles. Interactive computer simulators, such as APPLESCAB, have been developed for instructional uses. The APPLESCAB simulator incorporates all the important components of the apple scab disease system, including fungicide treatments, weather conditions, pathogen populations and levels of fungicide resistance, tree growth and development, and economic parameters such as cost of the chemical treatments and market value of the crop. By varying the different components, the simulator can demonstrate their effect on disease development and can predict the economic result of the epidemic.

***Agaricus*: Structure and life cycle:**

Introduction

The genus *Agaricus* comprises a group of 200 species which are heterotrophic, true nucleated, multicellular, macroscopic and fleshy in nature. This group includes fungi whose fruiting bodies are commonly known as mushrooms. The umbrella shaped mushrooms (which are edible), toadstools (which are non-edible or poisonous) beautify this earth with their presence in almost all seasons except the extreme summers and winters. The fruiting bodies of some species appear in early spring and disappear in summers, others appear only in rainy season and some appear sporadically whenever moisture is available. Hence the moist season (rainy weather) is the best season to explore, observe and collect the most beautiful, colourful (earthy flowers) fruiting bodies of mushrooms in hills as well as in plains.

The word mykos (from which the term Mycology is derived) means mushrooms and etymologically, mycology is the study of mushrooms. This group has been extensively studied in detail because man collected the fruiting bodies of mushrooms from diverse habitats to consume them as food, medicines etc.



Taxonomy

Alexopoulos C. J. Mims , C. W. and Blackwell, M.(1996) :

Kingdom : Fungi

Phylum : Basidiomycota

Class : Basidiomycetes

Order : Agaricales

Habit and Habitat

Agaricus is a multicellular, macroscopic, mostly saprophytic corticolous, parasitic and mycorrhizal in nature. Agarics are cosmopolitan in nature and occur in a wide variety of habitats ranging from the arctic to the tropics. Most of the species do show the preference to the diverse kind of substrata for example :-

□ In upland woody forests on decaying wood (lignicolous), decaying fallen leaves (foliicolous) eg. *A. lanipes*, *A. langei*, on moist soils (terrestrial), compost and dung (coprophylous) eg. *A. bisporus*, *A. bitorquis*.



Somatic structure (vegetative thallus)

The fruiting bodies (sporophores) of mushrooms as seen on the surface of substratum by the naked eye do not exhibit the complete fungal thallus but these are the sexual reproductive bodies bearing meiotic basidiospores on the part of mycelium.

The somatic structure or vegetative thallus of *Agaricus* consists of the mycelium (mass of hyphae) which is septate, thin walled, hyaline typically a basidiomycetous type. Here it arises as a primary mycelium from the monokaryotic basidiospores, which later become dikaryotic and tertiary mycelium.

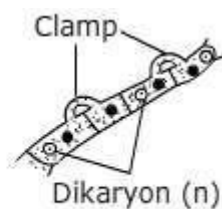
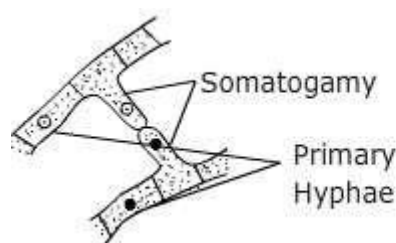
The mycelium of *Agaricus* is of three kinds:

Primary mycelium :The monokaryotic, haploid, basidiospore of (+) and (-) strain germinates immediately after dispersal on the substratum or on the moist soil (at 10- 15 degree temperature, 80—90% relative humidity, and 5.8—6.0 ph) by absorbing mineral nutrients directly in soluble form to give rise a germling or primary hyphal initial. This hyphal initial cell undergoes mitotic

division to form monokaryotic, uninucleated, haploid, hyaline, thinwalled primary mycelium which has a very short life in nature.

Secondary mycelium: The primary hyphae or monokaryotic hyphae with (+) or (-) strain haploid nuclei come in contact with each other to form dikaryotic or heterokaryotic mycelium by two methods :-

- i. **Somatogamy:** The somatic cells (vegetative cells) of primary or monokaryotic hyphae of opposite strains (+ and -) when they come in contact with each other directly on the substratum , the cell wall between the two cells (+ and-) dissolves , fusion of two protoplasts (plasmogamy) takes place, two nuclei (+and-) lie near each other to form a dikaryotic, haploid cell (somatogamy) . This dikaryotic (n) cell undergoes multiple mitotic divisions to form thin walled, multicellular, branched secondary mycelium. Each cell of this mycelium carry two genetically distinct (compatible) nuclei (+ and-) and is strictly heterokaryotic, dikaryon. fig.
- ii. **Clamp connection:** This method is very common to develop heterokaryotic, secondary mycelium from primary mycelium of opposite mating types. In this process the two compatible cells of opposite strain (+&-) of monokaryotic hyphae come in contact with each other, the cell wall between them dissolves, nuclear migration takes place . The nucleus of this dikaryotic cell divides mitotically and the daughter nucleus migrates in the adjacent cell with the help of a characteristic structure or a connection i.e. a short appressed hyphal branch which bypasses the transverse septum and connects the two adjacent daughter cells laterally called Clamp (or buckle) connection. It is a method which ensures that when a dikaryotic mycelium divides, each cell of these hyphae will contain two genetically distinct nuclei.



Tertiary mycelium : The dikaryotic, secondary mycelium carrying opposite strain(+and-) haploid nuclei in the hyphal cells branch profusely by mitosis and the hyphal branches anastomose with each other to form a complex tissue of fruiting body (basidiocarp).

Vegetative or hyphal cell

The *Agaricus* hyphal cell consists of outer cell wall (chitin microfibrils and glucan), inner plasma membrane (selectively permeable), true nucleus, haploid of + and - strain or both, cytoplasm, mitochondria, ribosomes, endoplasmic reticulum, golgi apparatus, small vacuoles and glycogen granules as reserve food material etc.

□ Structure of septum: The septum (the intersecting wall between each hyphal cell of septate hypha) in each cell of monokaryotic and dikaryotic hyphae of *Agaricus* is pierced by a narrow 0.1- 0.2 μ m width which is surrounded by a barrel called septal swelling. This kind of septal pore is known as Dolipore septum. This pore is overarched with a perforated cap i.e an extension of endoplasmic reticulum. This perforated cap is called parenthesome or pore cap which has several pores in it from which organelles like mitochondria can pass.

Reproduction

Asexual reproduction

The species of *Agaricus* undergo asexual reproduction very rarely. It produces two types of asexual spores i.e. oidium (thin walled, fragmented spore), and chlamydospores (thick walled, resting spore).

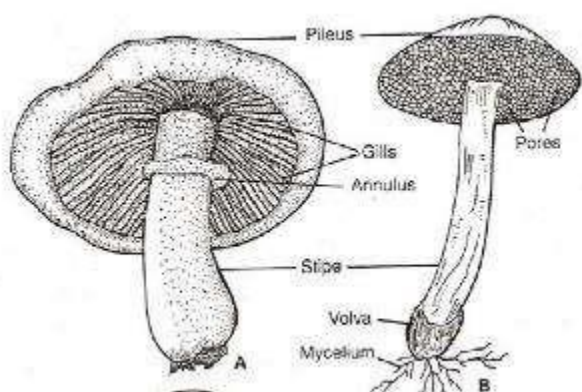
Oidium or fragmented spore : Oidia or arthrospores are thin walled, hyaline, fragmented, spores which are produced by the dissolution of middle lamellar wall between the adjacent cells on the aerial branches of monokaryotic and dikaryotic hyphae. The monokaryotic oidium fuses with the hyphal cell of the opposite strain to form the secondary heterokaryotic mycelium .The oidia of dikaryotic type germinate directly to give rise heterokaryotic mycelium eg. *Coprinus lagopus*. These type of spores are produced during favourable conditions i.e temperature 10 - 15°C , relative humidity 80-90% and pH 6-6.5 .

Chlamydospores or resting spores: It is a thick walled resting spore or perennating spore (seed like) produced during unfavourable conditions (drying of substratum, rise in temperature, and change in pH). The hyphal cells lose extra moisture content, glycogen particles (reserve food) get converted into fat droplets, cell wall becomes thickened by chitin deposition. These spores produced in chains are initially light coloured but later become dark brown, get separated from the rest of the parental mycelium

Sexual reproduction *Agaricus* does not have well defined sex organs or sexual gametes. The somatic or vegetative cells of primary hyphae of opposite mating types (+&- strains) function as sexual gametes. The two somatic cells of haploid primary hyphae of opposite strains (+&-) come in contact with each other by a process of somatogamy, oidization or by clamp connection and fusion of protoplasts (plasmogamy) bring two compatible nuclei (+&-) near each other. Homothallism is rare in agaricales eg. (*Coprinus sterculinus*). Most of the species exhibit either unifactorial or bifactorial homothallism . The two significant steps that is karyogamy (nuclear fusion of opposite mating +&- strains) and meiosis (reduction division) take place in the basidial cells of fertile layer (hymenium) of fruiting body i.e. basidiocarp . The basidiospores (meiotic) are produced on the probasidial cells exogenously at the abaxial(ventral) surface of the basidiocarp

Basidiocarp

The fruiting body or the basidiocarp formation is regulated or influenced by the interaction of multiple environmental factors:



Light: Diffused light .

Relative humidity : 80-90 % .

Temperature : 12-18°C .

pH of the substratum : 5.8-6.0 .

Mineral nutrients (ammonium salts etc.) and aeration.

These conditions favour the formation of fruiting bodies or the basidiocarp on the substratum.

The initial steps in fruiting body formation are :

1. An intricate hyphal lattice is formed by the interaction of hyphal branches .

2. From this hyphal lattice the bunch of aerial hyphae appear which eventually produce a round aggregation of tightly interwoven hyphae to form a button or a bud like structure, creamish white in colour called bud primordium (first basidiocarp stage) on the substratum.
3. The bud primordium grows in size and as it reaches 1.0 mm in diameter , a presumptive stipe, hymenium (fertile layer) and terminal pileus (cap) get differentiated simultaneously .
4. The elongation of stipe, expansion of terminal pileus in to upper (adaxial)smooth and lower(abaxial)infolded gilled lamellar structure and production of basidiospores occur quite rapidly and dramatically in the life cycle of Agaricus .

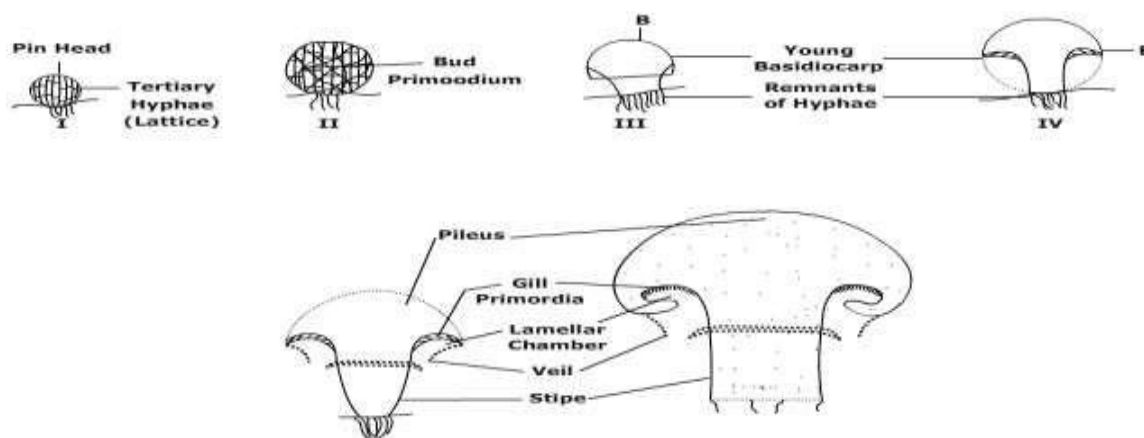


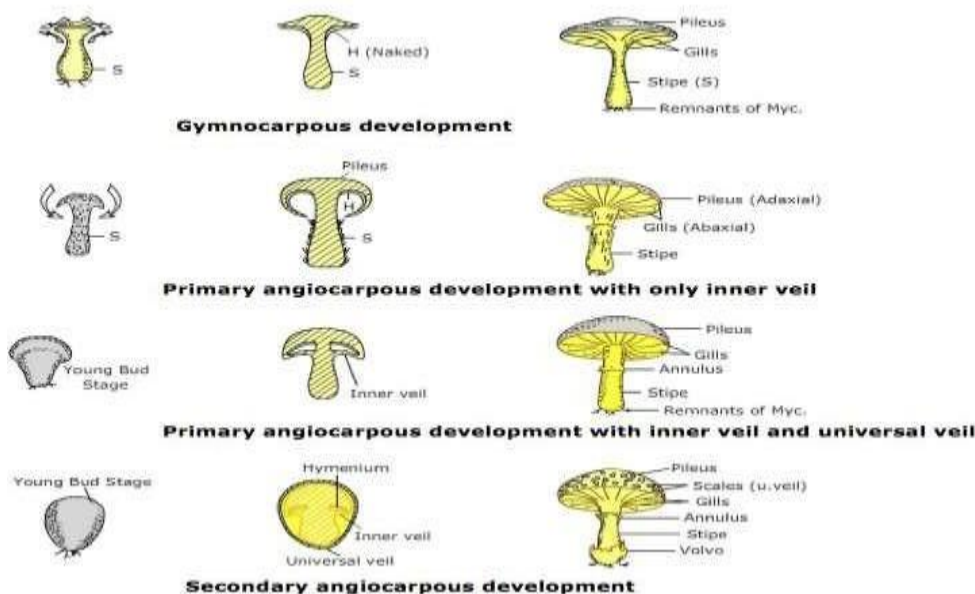
Figure: Diagrammatic representation of development of basidiocarp from pin head to mature fruiting body.

Hemiangiocarpous development: The hemiangiocarpous basidiocarp development is characterized by the following facts:

- The hymenium or fertile layer is enclosed by tissues (tertiary mycelium) of the basidiocarp even during early stages of its development .
- The margin of the pileus is connected to the basal stipe by a membrane called inner veil .
- The hymenium remains enclosed till the terminal cap (pileus) expands and matures .
- The veil tears from the margin of the pileus shortly before the spores mature and remains attached to the stipe as a ring or annulus .
- (In some species the veil tears in such a manner that it hangs down from the cap(pileus) like a thin cobwebby curtain called Cortina and in others the entire primordium is covered by a universal veil. This universal veil breaks and leaves a cup shaped body around and bulbous base

of the stipe. The remains of the parts of universal veil are seen as scales on the cap. These features help in classification and identification of many species as this feature is evanescent in other species).

Gymnocarpous and Pseudoangiocarpous development : In these two methods the hymenial (fertile) layer is formed initially on the outside of the fruiting (basidiocarp) body. In gymnocarpous development the hymenial tissue is not covered and remains naked till maturity. In pseudoangiocarpous method the hymenial tissue later becomes enclosed by the development of the outgrowth of the incurving margin of pileus and the stipe. The hymenium remains enclosed till the basidiocarp matures and the pileus expands to reveal the hymenial tissue .In these methods the vestigial structures are not seen at the maturity of the fruiting bodies (basidiocarp).



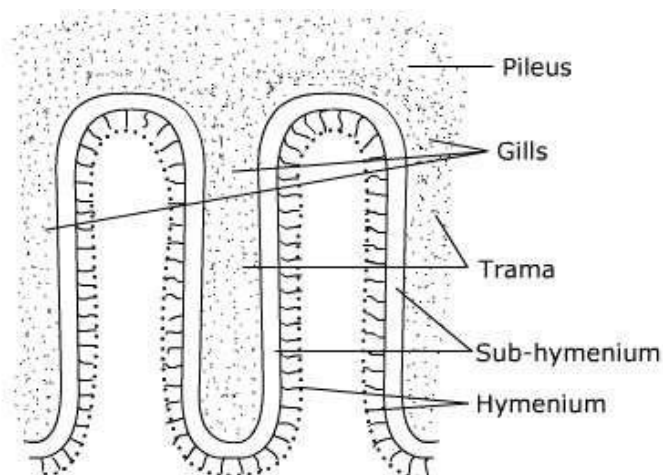
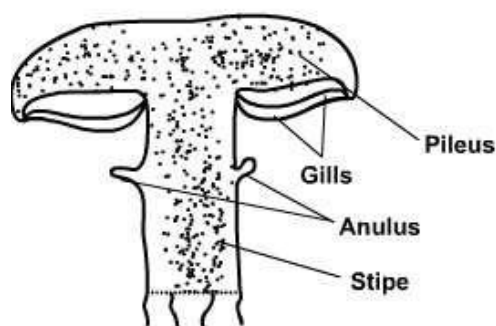
Structure of Basidiocarp: The basidiocarp of *Agaricus* which bears basidiospores consists of the following tissues:

- Hymenium:** The fertile layer of anastomosed secondary hyphae is found beneath the pileus (terminal cap). It lines the gills or lamellae exogenously which hang below in the ventral (abaxial) surface of the pileus.
- Gills:** The gills are the thin strips of tissues which radiate from the margin of the pileus in towards the stipe. The structure and position of the inner edge of gills towards stipe is a valuable

and interesting feature in many agaricales for taxonomists. The gills in some species are free from stalk while in others they are attached directly to the stipe called adnate. The decurrent gills are attached and run down the stipe for some distance.

c) Trama : The trama (L. Trama-Woof) is the inner most tissue which lie in the center of the gills. It is an important feature of taxonomic interest Singer (1975). The central tissue (trama) is of two types:

- The trama consists of plectenchymatous tissue which is made up of elongated hyphae.
- The trama tissue contains large, globose or oval cells called sphaerocysts scattered along the elongated hyphal cells (eg. *Russula* and *Lactarius*). The trama is surrounded on both sides by the fertile layer (hymenium) in which closely packed layer of basidia are interspersed with basidioles or cystidia or both in some species . The cystidia are long and extend from one gill to the next to keep the gills apart from each other (*Coprinus*) .



Basidium : The basidium is a cell or a structure found at the terminal end of dikaryotic hypha in the hymenial layer of the gill exogenously. In the basidial cell the dikaryotic nuclei fuse with each other (karyogamy) to form diploid nucleus ($2n$) which immediately undergoes meiosis (reduction division) to form a definite number of basidiospores (n). Each basidial cell bears a definite number of haploid basidiospores (2 or 4) which at maturity are forcibly shot off to disperse and fall below the pileus under the influence of gravity on the substratum. In still air thesehaploid basidiospores (+ and-) get deposited below the pileus in a mass to form the spore print.

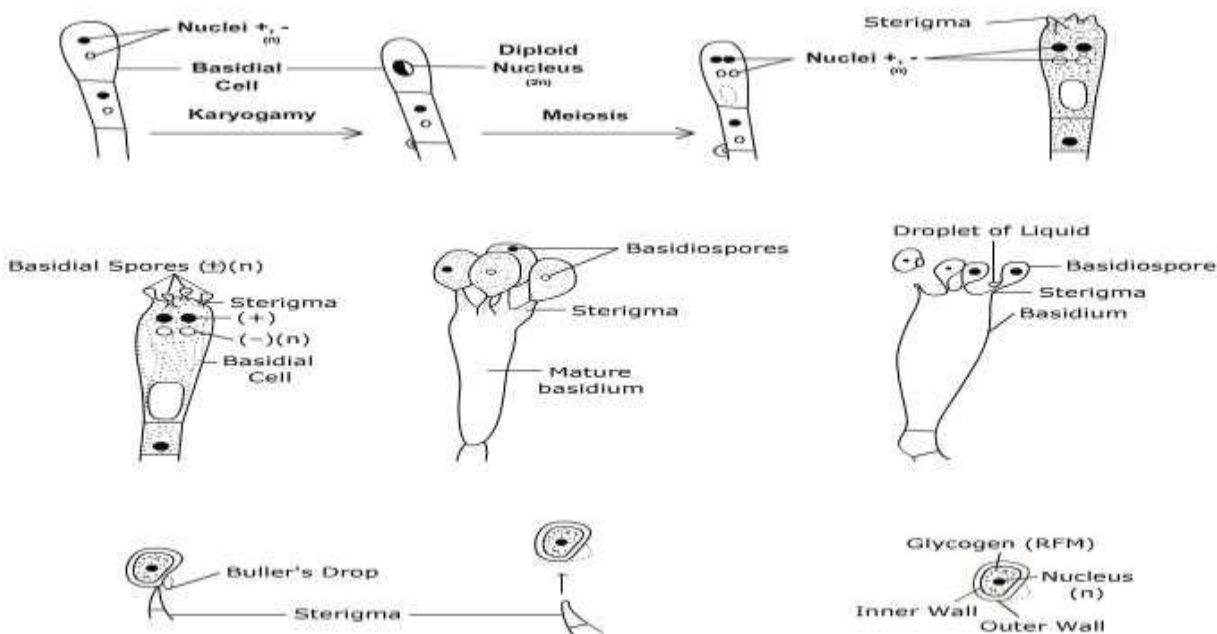


Figure: Development of basidium and formation of spores

Basidiospore: The haploid, monokaryotic basidiospore varies in size, shape, colour and is thick walled and multilayered (3-5), eg. three layered in *Psilocybe sp.* and *Agaricus brunnescens (bisporus)*. It consists of protoplasm, haploid nucleus (+or -), ribosomes, mitochondria, lipid bodies, vacuoles, glycogen granules as reserve food material . The basidiospores exhibit distinct colours eg. in *Chlorophyllum molybdites* spores are green, in *Russula sp.* the spores are yellow and in other species spores are white, pink, brown, black and smoky-grey. The spore colour is not always the same as that of gills and is determined by making a spore print on white and black paper .

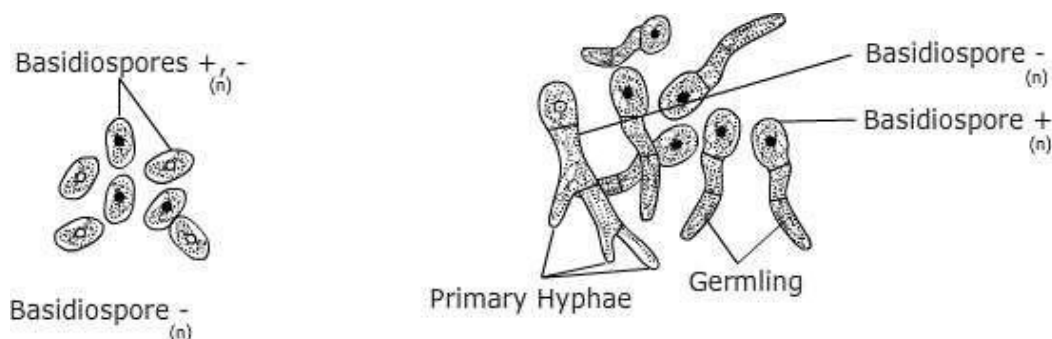


Figure (above): Basidiospores and formation primary hyphae.

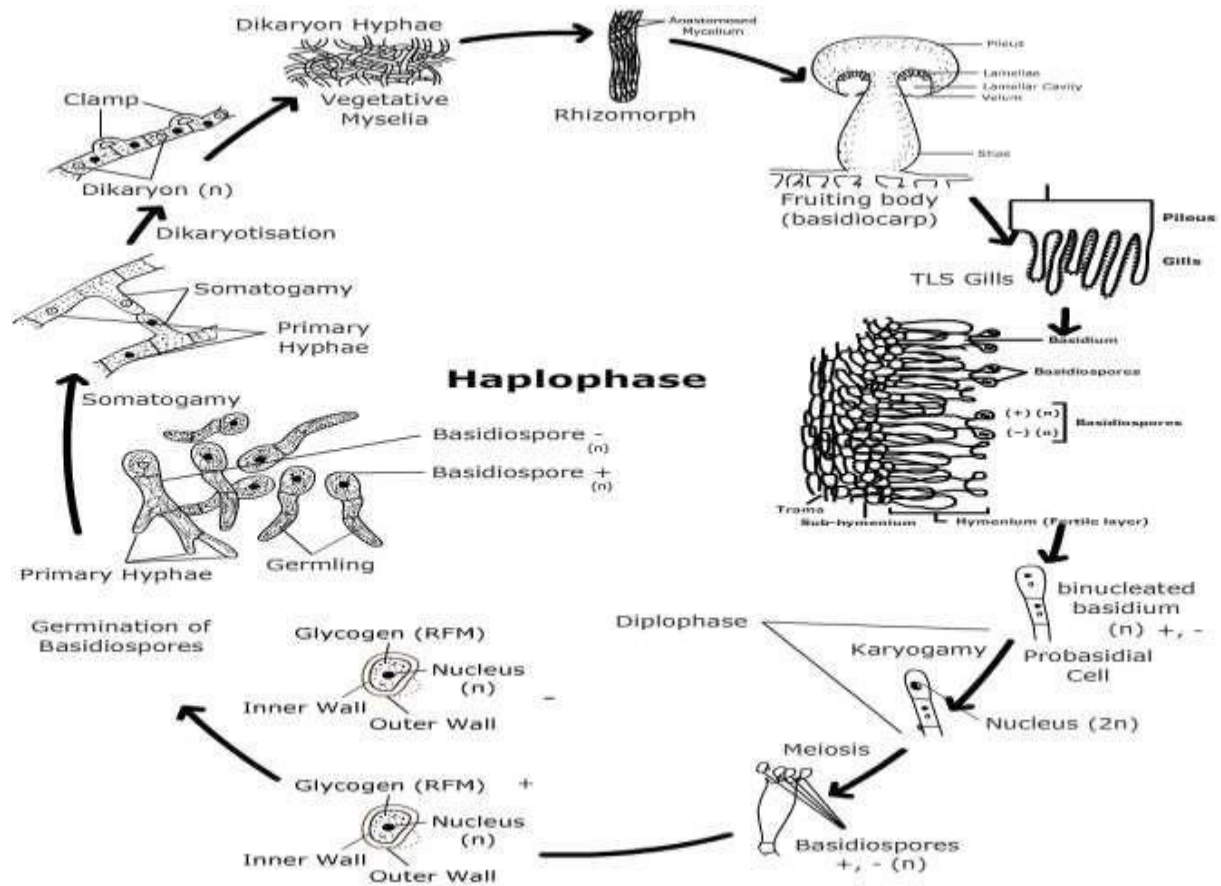


Figure: Diagrammatic life cycle of *Agaricus* sp.

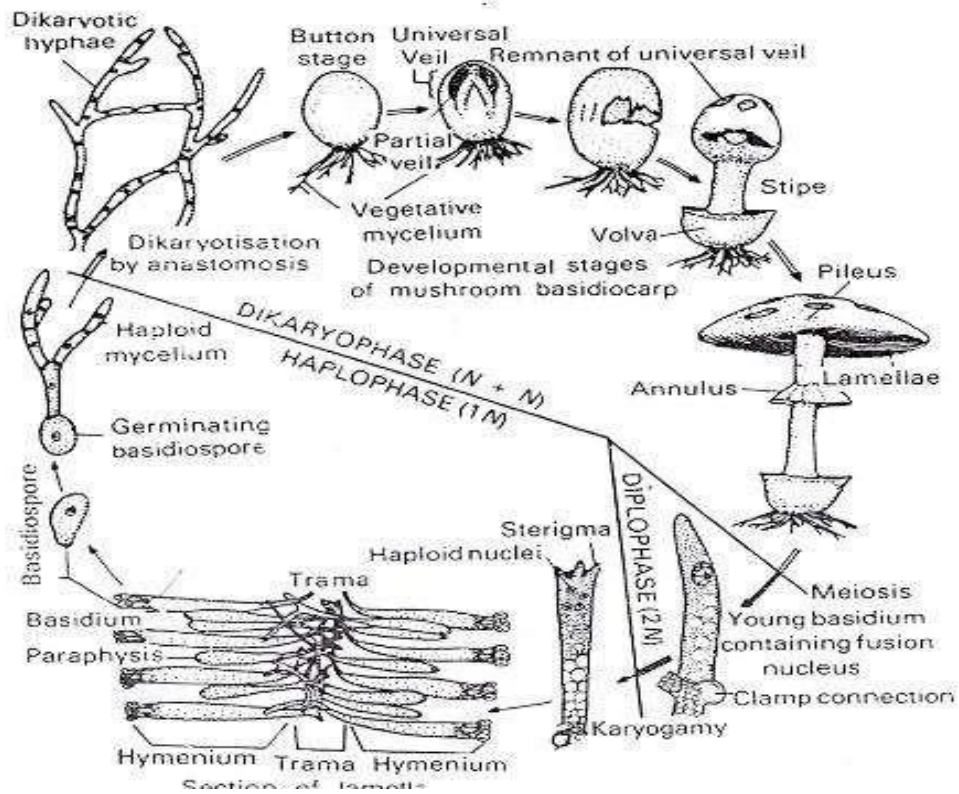


Figure: Life cycle of a typical agaric (*Amanita* sp.)