Puccinia

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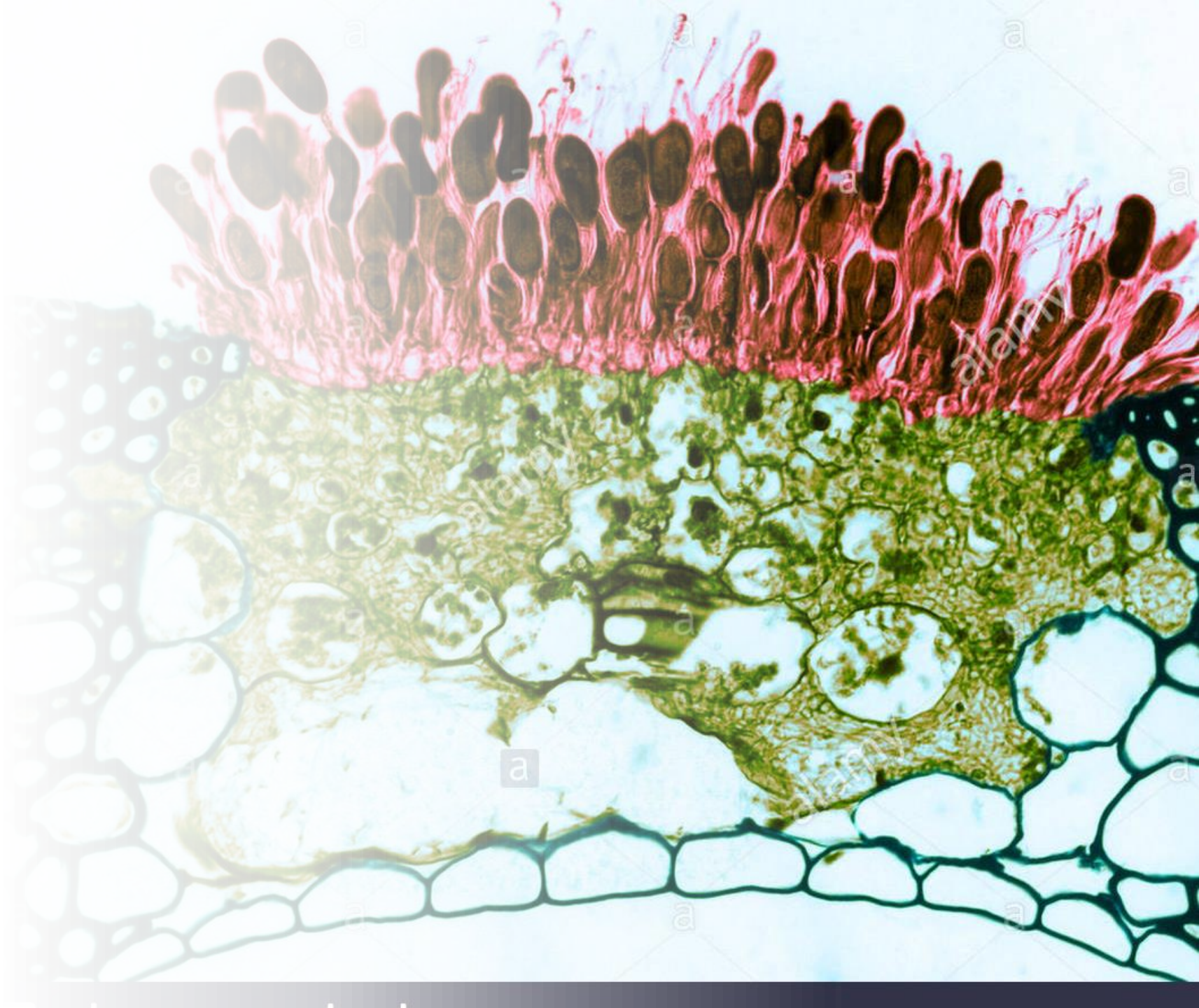


Systematic position

- Kingdom: Mycota
- Division: Eumycotina
- Class: Basidiomycetes
- Sub-Class: Heterobasidiomycetes (Teliomycetidae)
- Order: Uredinales
- Family: Pucciniaceae
- Genus: ***Puccinia***

Habit and Habitat

- *Puccinia* grows as an **obligate parasite** on vascular plants especially on wheat and other cereals.
- Most of the species are **polymorphic (produce more than one type of spores)**, **heteroecious (require two host for the completion of their life cycle).**
- Only a few species e.g**. *P. helianthi* and *P. butleri* are autoecious**
- In India wheat rust appears by the **end of November in Southern part and in March in Northern part.**
- The name of genus has been kept in the honour of great Indian anatomist**, T. Puccinia**

Important diseases caused by Puccinia

- Puccinia infects a number of host and causes popularity known **rust disease**.
- The **loose cluster of spores, called sori** are produced on the leaves and stems which may be orange or red in colour giving the characteristic rust like appearance to the diseased host.
- Pucciniaceae family is characterized by stalked **teleutospores**

1. ***Puccinia graminis*** – Black rust of wheat
2. ***P. striiformis-*** Yellow rust of wheat
3. ***P. sacchari-*** leaf rust of Sugarcane
4. ***P. sorghi-*** Crown rust of Jowar

Disease Symptoms of Wheat Rust

Black Rust of Wheat

- This disease is caused by the fungus ***P. gramnis tritici***
- The disease is called **black stem rust** because the fungus develops black rusty symptoms on wheat
- The fungus is **polymorphic, macrocyclic** and **heteroecious**
- **Uredopostules** are **oblong, vertically elongated, reddish brown and granular.** Postules are also called **uredosori** or **uredia** since they contain uredospores. Postules develop sub-epidermis exposing the powdery mass of reddish-brown uredospores giving the rusty appearance.
- **Teleutopustules** are produced at the end of season when atmosphere becomes hot and dry. These pustules are also called **teleutosori or tellia** since they contain teleutosores. Teleutosori are black or dark drown, irregular, elongated and crust like pustules which develop on the stem, leafsheaths and clums of wheat plants
- Its secondary host is **Barberry plant**

Yellow or Stripe Rust of Wheat

- This disease is caused by the fungus *P. striformis (P. glumarum*) in only northern and eastern parts in the month of January.
- The **rust pustules (Uredosori**) and small, oval, Lemon yellow coloured and arranged in linear fashion, hence the name **stripe rust.**
- The pustules are confined to the leaves
- Uredosori are sub-epidermal in early stage but later on rupture through the epidermis. They remain quite close to each other but do not fuse.
- **Teleutosori** are dull black in colour and arranged linearly. The teleutosori do not break through the epidermis as in black rust and remain as flat black crusts
- Its alternate host is ***Bromus japonicas***

Brown or orange Rust of Wheat

- Brown or orange rust disease of wheat is caused by the fungus *P. triticina or (P. recondite)*
- It appears by the end of November in Bihar
- Uredopustules are brown or orange coloured, scattered irregularly and when burst open, the ureodspores are released.
- The pustules occur exclusively on the leaves and hence is called leaf rust.
- **Teleutosori** are of **rare occurrence**. They are linear to oval in outline, black coloured and divided into small groups by appearance of paraphyses. Teleutosori do not burst through the epidermis.
- Its alternate host is ***Thallictrum flavum*.**

Vegetative structure

Mycelium

- Mycelium is the **somatic or vegetative structure of the fungus.**
- It is found intercellularly or intracellularly.
- Mycelium is about 5.5µm in diameter with septa developing at long intervals.
- The hyphal cells may be uninucleate or binucleate depending upon the stage in life cycle.
- It is **monokaryotic in its first phase and dikaryotic in later stages.**
- **Mycelium is well developed, branched septate and contains haustoria.**
- Haustoria may be small, rounded or globular or connections are found in dikaryotic mycelium which are not very frequent.
- The monokaryotic or primary mycelium is found on the alternate or secondary host, e.g., **Barberry leaf in case of black rust.**
- **The dikaryotic or secondary mycelium is found on the primary host** e.g., wheat in case of black, yellow or orange rusts.

Reproductive Structure

Study of Spores

- ***P. graminis tritici*** is a **Macrocyclic Fungi.** It produces **five types of spores**
- They are **pycniospores, aeciospores, uredospores, teleutospores** and **basidiospores.**
- Out of theses five types of spores **pycniospores and aeciospores** are found on **alternate or secondary host i.e., barberry leaf** and remaining three types of spores are found on primary host i.e. **wheat.**

Five types of spore in *P. graminis tritici*

Stage 0: Spermagonium or **Pycnidial stage** (Uninucleate pycnidiospores)

Stage 1: Aecium or **Aecidial stage** (Binucleate Aecidiospores)

Stage 2: Uredium or **Uredial stage** (Binucleate urediospores)

Stage 3: Telium or **Teleutostage** (Binucleate Teleutospores)

Stage 4: Basidium or **Basidial stage (**Uninucleate Basidiospores)

Uredo Stage

- **Stage on wheat rust- Uredo** and teleutostages develop on **wheat plant.**
- The **mycelium** is the host tissue is **dikaryotic and branched**; hyphae septate and present in the **intercellular spaces.**
- A **cluster of uredospores** is seen projecting out through ruptured epidermis.
- The **uredospores** are attached to the underlying mycelium with the help of stalks, i.e. uredospores are produced from this underlying mycelium.
- Each uredospore **is binucleate, stalked, oval or rounded or oblong in shape** and possess a **double layered thick wall.**
- **The outer wall layer**, called **exosporium or exine**, is thick brownish, echinulate or variously sculptured in ***P. graminis tritici*** and hyaline in ***P. stniformis (P. glomarus***) whereas the inner wall layer called intine in **thin, smooth and membranous.**
- Each uredospore possesses one to four germ pores in P. *graminis tritici*, 6-10 in *Puccinia striiformis* and about six *in P. triticina*

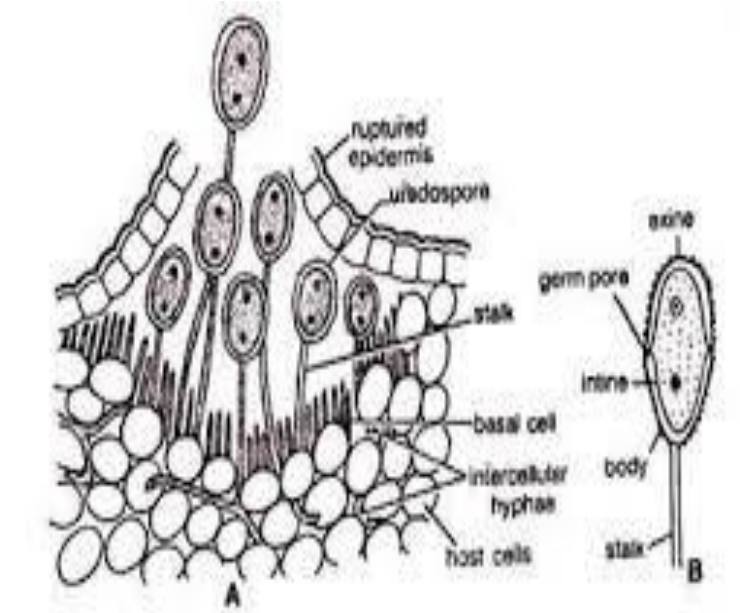
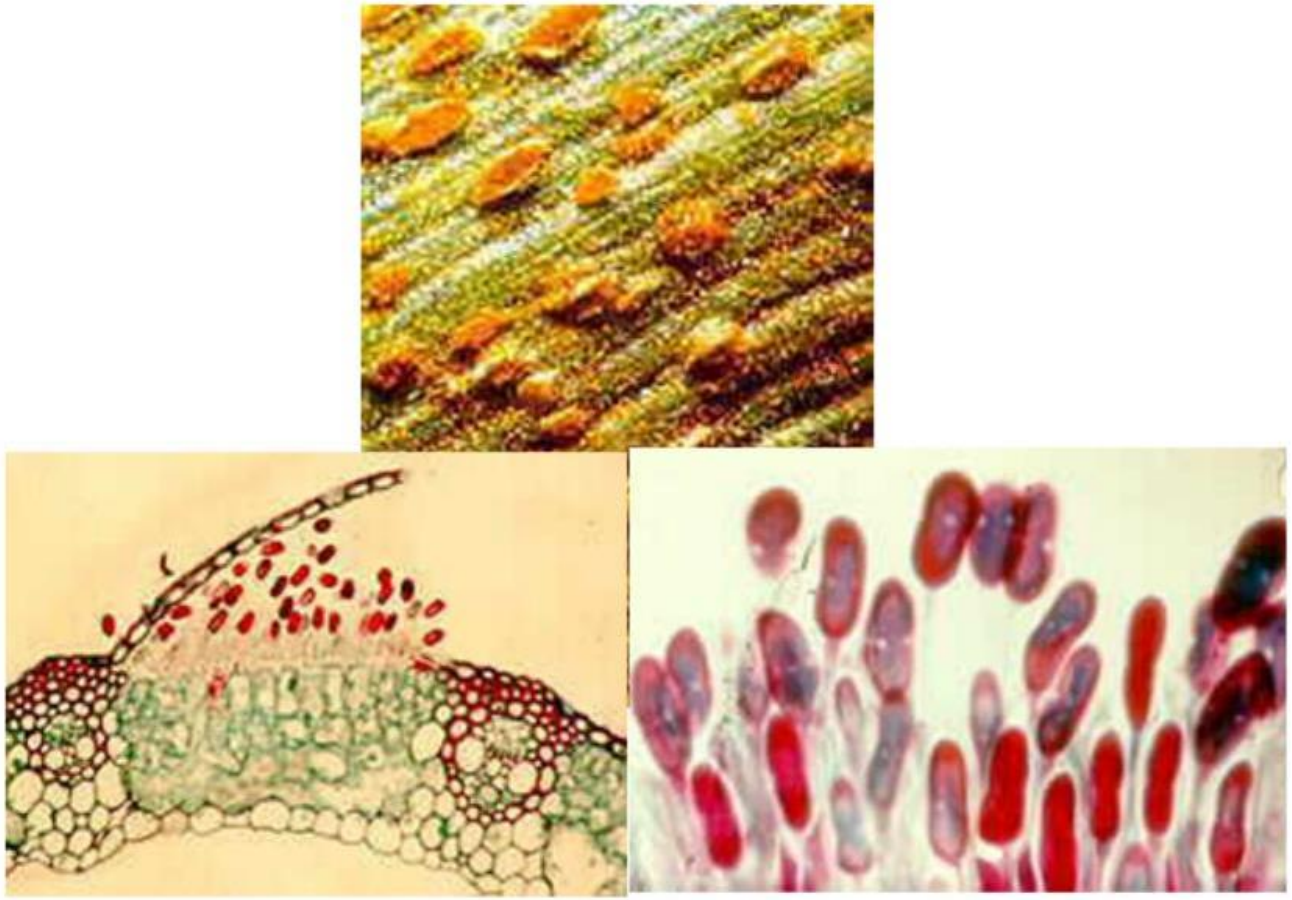


Fig. 4 (A–B). *Puccinia graminis* : T.S. wheat leaf passing through uredosorus, (B) A uredospore

Development of Uredosori and Uredospores

- The development of the uredosori begins with the germination of **uredospores or aecidiospores**.
- In presence of **sufficient moisture**, these **spores develop germ tubes** which grow over the host epidermis for some time and penetrate the host through stoma.
- The germ tube forms an **inflated vesicle like appresorium**.
- A **narrow infection hypha,** now develops from appresorium, grows and expands in substomatal vesicle.
- The vesicle ultimately gives rise to much **branched sepatate mycelium which penetrates the intercellular spaces of the host cells in subepidermal regions.** The mycelium now begins to absorb nutrition from host cells through **haustoria.**
- At each point of infection, **the mycelium collects to form a compact mass of cells beneath the epidermis**.
- A layer of parallel binucleate cells called **basal cells arise from this mass and presses the host epidermis from below.**
- The basal cell divides transversely into a lower, **sterile foot cell** an**d an upper cell.** The upper cell again divides transversely into stalk cell that elongates and mature within 10-12 days.

Uredia and Uredospores



Uredia

- As the uredospores mature and their **stalk elongate**, the host epidermis lying above them is pressed and ultimately it ruptures.
- The **uredospores are easily detached and accumulate in a reddish brown granular powdery mass over the infected leaf.**
- If the conditions are **favourable (cloudy weather)** several successive crops of uredospores are produced in a season.
- Even a single sourous may produce uredospore for a long time since new basal cells are produced continuously.

Teleuto stage

- By the end of the season when the atmosphere become **hot** and **dry,** the **dikaryotic mycelium** which had been producing uredospores, now begins to form **dark coloured teleutospores**.
- Sometimes the uredosorus may also start **producing teleutospores**.
- This switch over from uredospore to teleutospore is probably governed by the photosynthetic activity of the host.

T.S. Wheat stem or V.S. leaf sheath passing through teleutosorus of *P. graminis tritici*

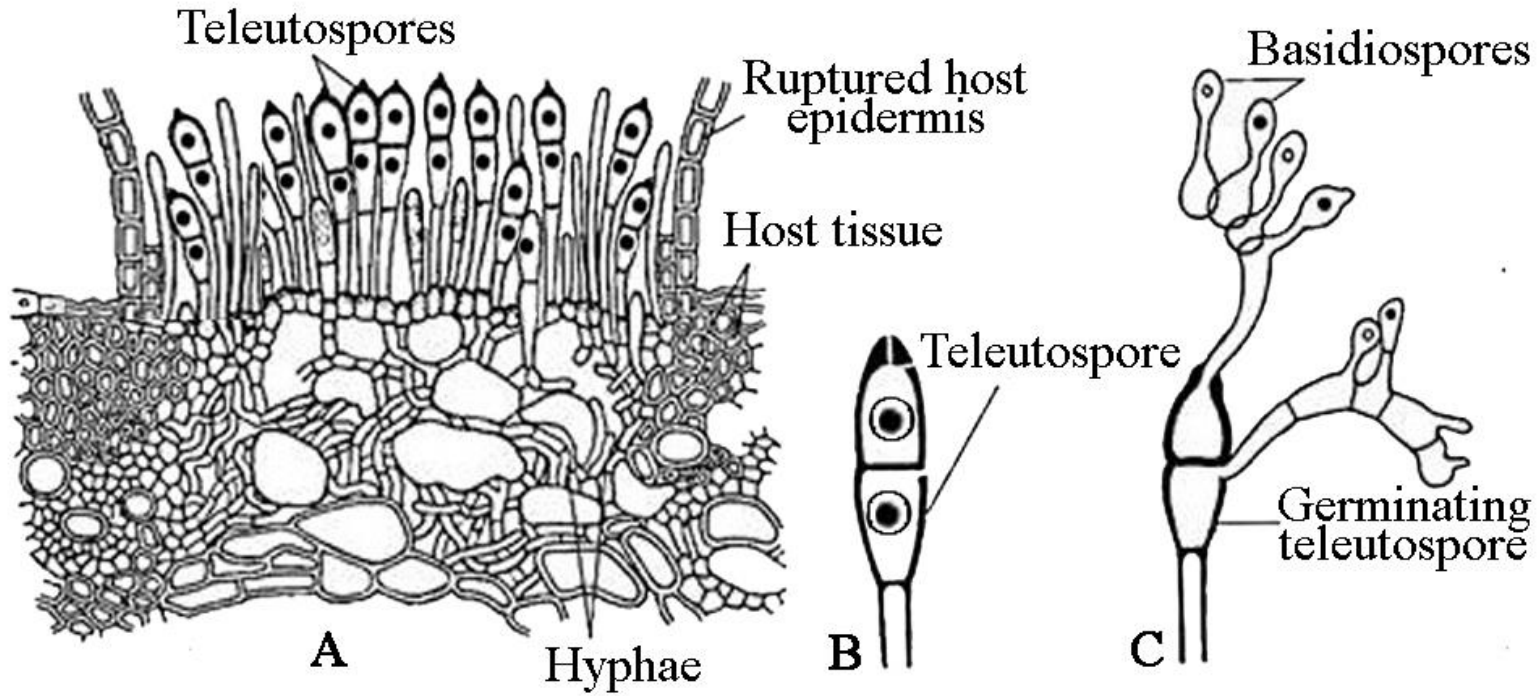


Fig: ***Puccinia graminis.*** **(A)** Section through teleutosorus showing teleutospores, **(B)** Single teleutospores, **(C)** Germinating teleutospore.

T.S. Wheat stem or V.S. leaf sheath passing through teleutosorus of *P. graminis tritici*

- The mycelium in host tissue is **dikaryotic** and **branched**; hyphae septate and present intercellularly. **Globular haustoria are also seen intracellularly**
- A cluster of teleutospores is seen projecting out through the ruptured epidermis or pustules.
- Teleutospores are produced from the same **inter-cellular mycelium which produces uredospores.**
- Each teleutospore is **elongated or spindle shaped, bicelled, long stalked, dark brown in colour quite thick and** bilayered.
- The **outer wall layer (exosporium)is thick, smooth, dark brown or black in color** and the **inner wall layer (endosporium or intine) is thin delicate and hyaline.**
- The **apex of each teleutospore is either rounded or pointed.**
- Each cell of a teleutospore has **dense cytoplasm, oil and a germ pore.** The upper cell has a narrow apical germ pore while the lower cell has lateral germ pore near to the septum.
- The cells of young **teleutospores are binucleate but at maturity both the nuclei fuse.** Thus, **each cell of mature teleutospore contains single diploid nucleus**.
- The teleutospores when detach from the primary host (wheat plant) they usually fall on the soil and germinate to produce basidia and basidiodpores which ultimately infect the alternate host.

Development of teleutosori and teleotospores

- The sorus containing teleutospores is called **teleutosorus**.
- The **teleutosori** appear as black, elongated pustules which develop on the leaf heaths as black, elongated pustules which develop on leaf sheaths and clums of wheat plant.
- The development of teleutospores is similar to uredospores except that **the upper daughter cell of the basal cell cuts off a row of three cell**s.
- The two upper cells form the **bicelled body of the spores** and the third lower cell elongates to become long stalk.
- The host epidermis overlying the teleutosori gets ruptured and teleutospores are exposed and detached.

Germination of teleutospores

- Teleutospores are **dormant spores** and therefore cannot germinate immediately.
- They require a **resting period of several months and help the fungus to survive until** next season.
- During germination, each cell of a teleutospore develops a tubular germ tube **celled promycelium or epibasidium.**
- The diploid nucleus now migrates into the promycelium and divides there by meiotic division to form **4 haploid nuclei.**
- Transverse walls are laid down in the promycelium which **becomes 4-celled, each cell with single nucleus.**
- Each cell of promycelium now develops a **slender, lateral outgrowth called sterigma** which bears **basidiospore at its tip.**
- Thus, a **promycelium form 4 basidiospores** out of which **two are of + type and two of – type**.
- The basidiospores are discharged forcibly by water droplet method disseminated by wind.
- On falling in a water drop, basidiospores germinate by giving out slender germ tube. **The germtube cannot reinfect the wheat plant but can infect the alternate host , Berberis and Mohanias.**

Alternate host barberry leaf showing symptoms

- The specimen of the infected **barberry *(Berberis vulgaris*)** leaf shows symptoms of pycnidial and aecidial stages of ***P. graminis tritici.***
- The barberry plants are tall shrub growing commonly in foothills e.g., in Nanital. The barberry plant acts as an alternate or secondary host of ***P. graminis tritici.***
- The leaves of barberry plant get **infected b**y the **germination of basidiospore**s on it.
- Yellowish specks are present on the upper (dorsal) surface of an infected leaf which indicate the presence of **underlying pycnidia or spermogoni**a
- The lower or ventral surface of infected leaf shows the presence of a cluster of small yellow coloured cup like outgrowths. These are actually **aecidial cups.**
- **Pycnidia develops from monokaryotic mycelium**
- **Aecidia develops from dikaryotic mycelium**
- Pycnidial cups produce **pycnidiospores** whereas aecidial cups produce **aecidiospores.**
- **Aecidiospores infect primary host (Wheat)**
- The leaf-shape is characteristic with distinct midrib and margin containing several pointed spines

Barberry: Alternate Host to *Puccinia graminis*

- Common shrub
- Introduced from Europe

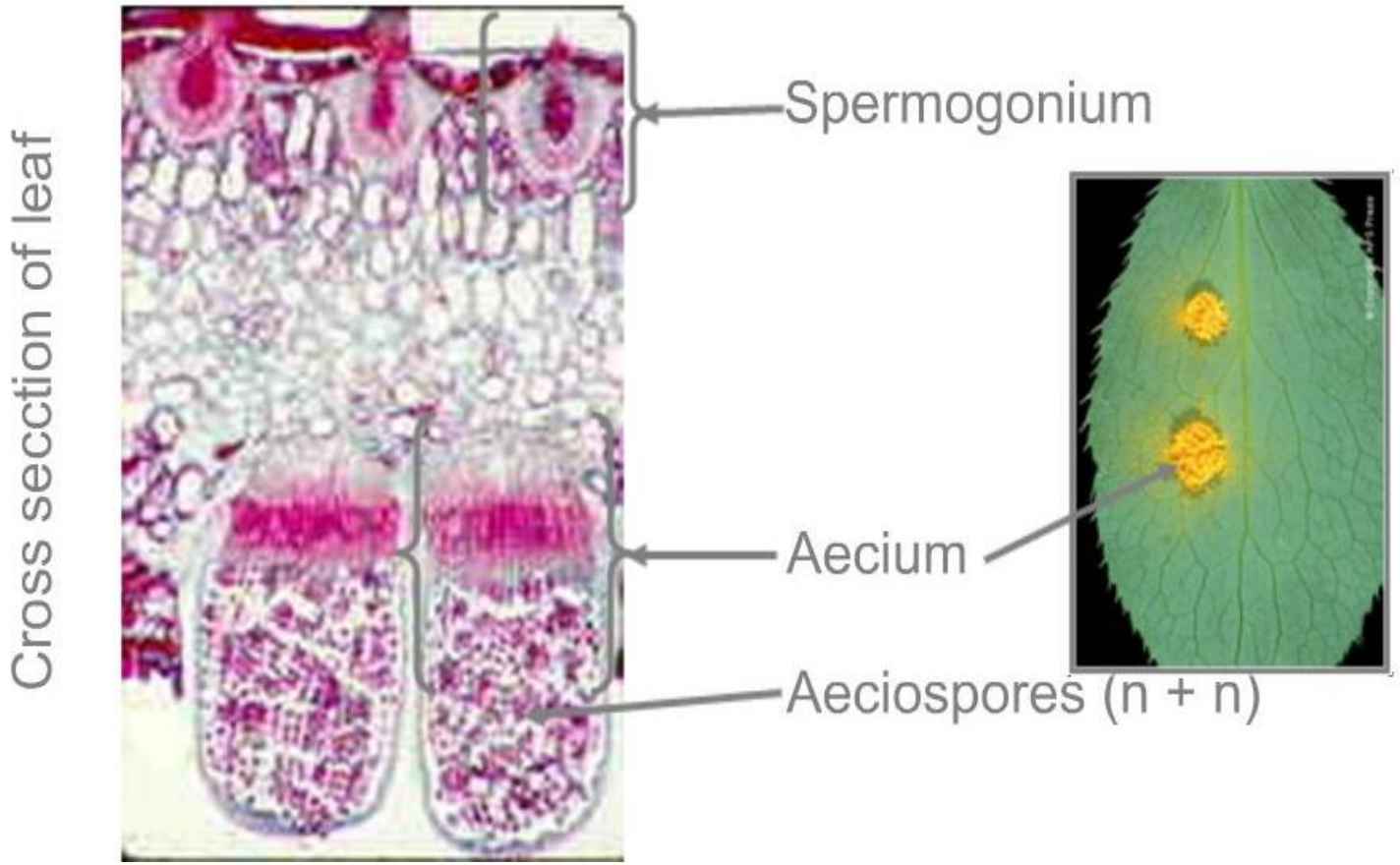
Barberry

Pycinidal or spermogonial stages

- When reaching the barberry host, the basidiospores germinate and their germ-tubes directly penetrates the epidermal cells of barberry leaves.
- Within 4-5 days of infection the primary mycelium accumulates in between upper epidermis and palisade cells of the leaf in form of dense mycelia mats.
- These mats are the primordial of **spermogonia or pycnidia**. The primordium is soon differentiated into oval to **flask shaped spermogonia**.
- The entire spermogonium remains embedded inside the leaf except the ostiole which projects above the surface.
- The region of **spermogonium adjacent to ostiole develops straight pointed hyphae, the periphyses and flexuous hyphae, the receptive hyphae**.
- Both these hyphae project outward, well beyond the ostiole. The bulbous or flask shaped cavity of the spermogonium is lined by a layer of numerous uninucleate elongated cells. These are **spores called spermatia or pycnidiospores.**
- The **spermatia soon detach from spermatophore** and are extruded through ostiole in a droplet of nectar like fluid secreted by the spermogonium.
- The **nectar drop usually covers the receptive hyphae and mass of spermatia just outside the ostiole**.
- A spermatium is more or less oval in shape with distinct large haploid nucleus and a little amount of cytoplasm.

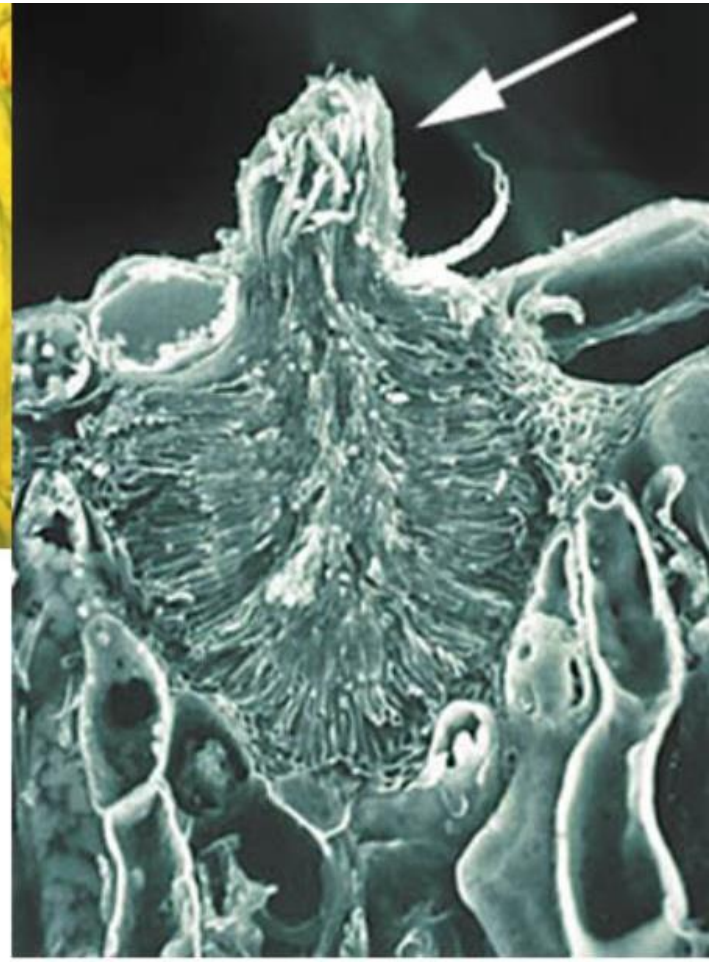
Pycnidial cup and Pycnidiospores

- **Pycnidial cup or pycnidium** is formed from the **monokaryotic mycelium** produced by the germination of each basidiospore.
- Pycnidia are present on the **dorsal or upper surface of l**eaf. They are **subepidermal , oval or flask-shaped structure, each with an opening apex, called ostiole.**
- The inner lining of the cup is called **peridium**. Its cells produce different types of outgrowth like **paraphysis, pycnidiophores or spermatiophores and receptive hyphae, in addition to periphyses.**
- The **paraphyses are sterile hyphae**, found intermixed with pycnidiophores inside the cup.
- **Periphyses** are found at the neck region of the ostiole and project towards outside through the ostiole.
- **Pycnidiophores** or **spermatiophores** are indefinite in number, club-shaped with flat broad end attached to the base of the cup and upper pointed or narrow end being free and present towards the centre of cavity.



Barberry Infection

- The terminal and narrow free end of **each spermatiophore** cuts numerous small, unicellular, oval or rounded, haploid structure called spermatia, or **pycnidiospores arranged in basigenous chains**.
- Receptive hyphae are quite long (longer than periphyses) **septate, branched or unbranched vertical and projecting out through the ostiole.**
- They are also called **flexuous hyphae.**
- The **pycnidiospore** almost fill up the cavity of the flask or **pycnidium.**
- The **spermatiospores or spermatia** behave like male sex cells and **receptive hyphae as female sex organs.**



Spermogonium
Spermatia
Receptive hyphae

Spermogonium

Spermatization

- It has been proved beyond doubt that a **spermogonium is either of + or – type (strain).**
- For the formation of next stage **(aecidial),** it is essential that spermatia of one strain must fuse with the receptive hyphae of the opposite strain.
- This exchange of spermatia takes place chiefly through flies which get attracted towards nectar or by the coalescence of nearly droplets or by rain.
- **Fusion of spermatia and receptive hyphae is termed as spermatization** and this results into the formation of **dikaryotic mycelium.**

Aceidial Stage

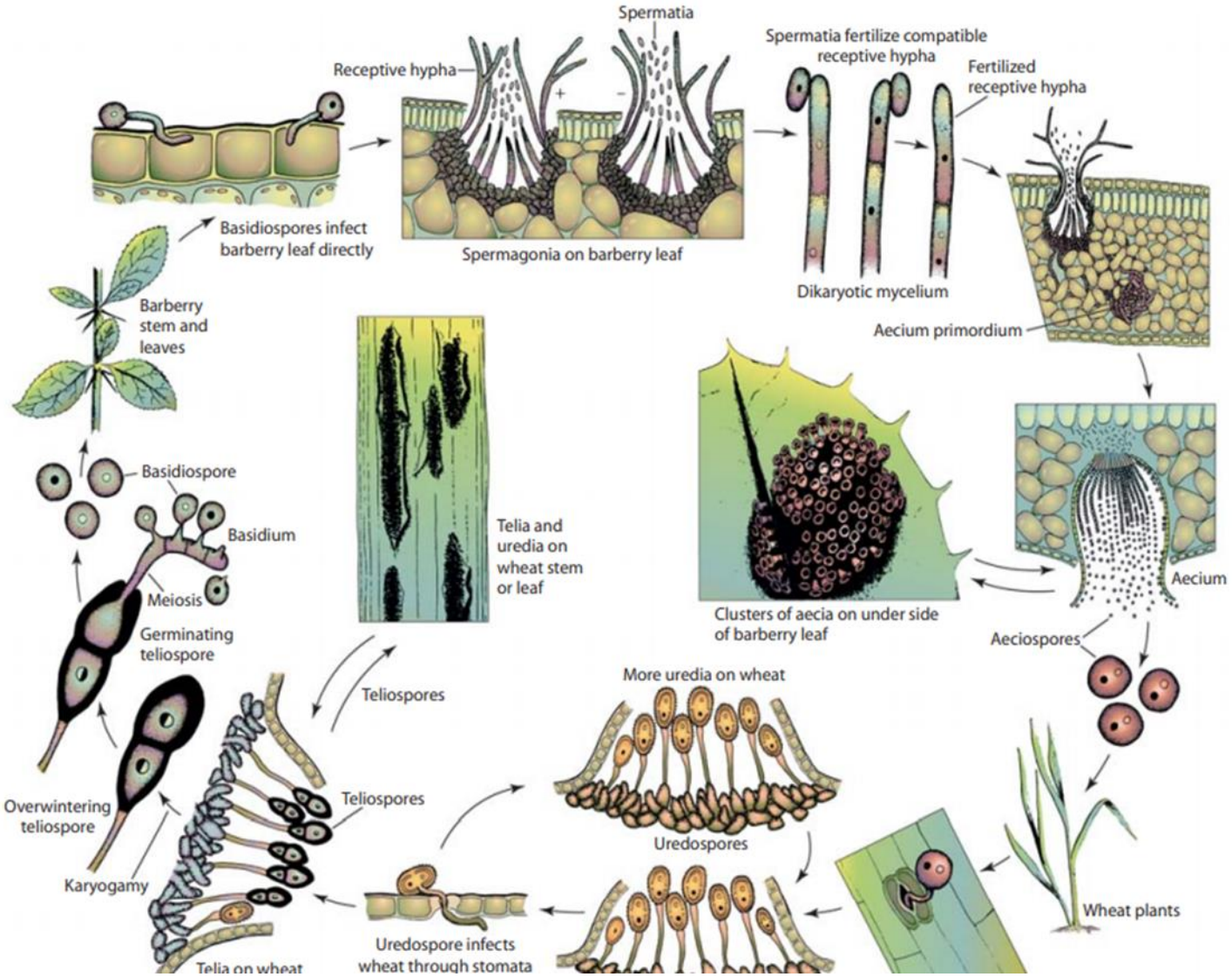
- The same primary mycelium **which formed spermogonia continues to grow down towards the lower epidermis of barberry leaf.**
- After reaching there the hyphae accumulate in the form of globose masses called **protoaecidia.**
- A **protoaecidium c**onsists of a plate like layer of basal cells and a mass of globose cells usually called displacement cells.
- The displacement cells face the lower epidermis and ultimately wither to form a cavity.
- Further **development of protoaecidium** is checked until spermatization has taken place. If there is no spermatization the protoaecidium may remain as such or may disintegrate.
- When spermatium of one strain is transferred to receptive hyphae of opposite strain, spermatization takes place.
- During this process **the spermatial contents passes from cell to cell through septal pore of the mycelium and ultimately reaches the basal cells of the protoaceidium making them dikaryotic mycelium is termed as dikaryotization or diploidization**, In this species dikaryotization has also been reported by fusion of primary hyphae within barbery leaf.

- Soon after dikaryotization , the protoaecidium matures into an aecidium containing aecidiospores.
- The **dikaryotized basal cells** are arranged in vertically elongated cells, the sporophores.
- Each sporophores cuts off a chain of binucleate cells.
- The chain of cells grows in the cavity formed by the disintegraton of displacement cell.
- The **chain alternately consists of aecidiospores and sterile intercalary dis**c. The disc integrates and the spores are discharged violently.
- A young aecidium is surrounded by a **protective layer called peridium, as the aecidium matures the peridial layer breaks, spreads out giving a bell shaped appearance**.
- An aecidium thus appears as a white cup filled with golden yellow mass of aecidiospores. More than 60 billion aecidiospores may be produced on a single barberry plant.
- **Aecidiospores are polygonial in shape** when inside the aecial cup but become spherical after discharge.
- Each spore is **thick walled, binucleate and has 6 germ pores**.
- They are **disseminated by wind and are capable of immediate germination**.
- The **aecidiospores cannot reinfect the barberry plant.**
- **When falling on wheat plant they germinate in water drop.**
- The germtubes infect the host through stomata and form uredospores within 10-12 days.
- This completes the life cycle of ***Puccinia graminis tritici.***

Aecium and aeciospores

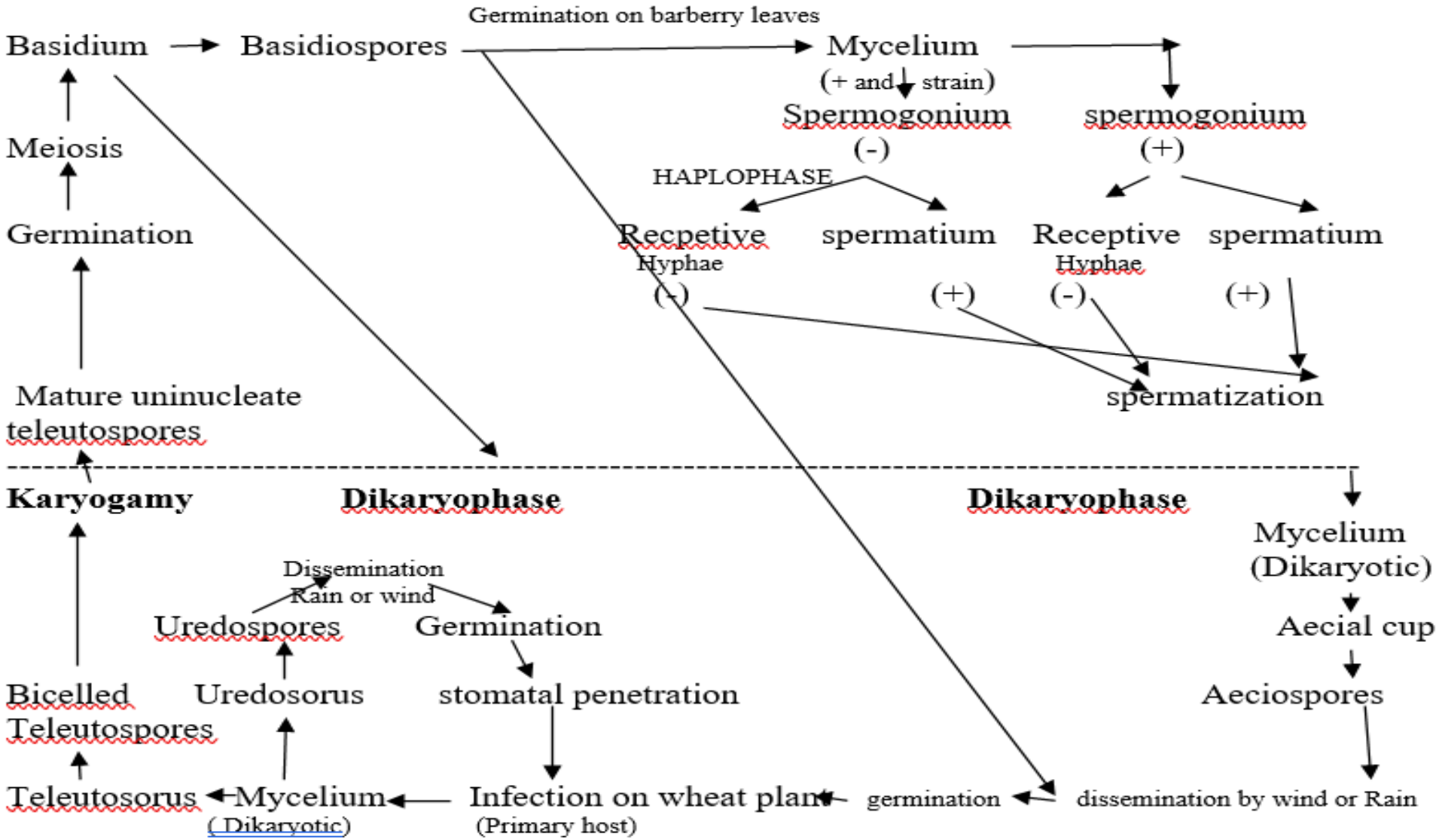
Aecidial cups and Aecidiospores

- The aecidial cups or aecidia are present on the lower (or ventral) surface of leaf.
- They develop from dikaryotic mycelium
- An **aecidium is a inverted cup-shaped structure**, surrounded by an inner linning peridium which is broken towards the lower side (base) due to pressure exerted by spores.
- From the base of the cup project out several palisade like arranged straight or curved large basal cells or sporophores.
- The tip of each basal cell cuts **off larger, unicellular, binucleate, rounded or multigonal (hexagonal) aecidiospores alternating with smaller disc-shaped disjuncture cells.**
- The basal aecidiospores look hexagonal due to compact arrangement but upper spores assume spherical shape and orange colour when become free.
- The aecidiospores are disseminated by dissolution of disjuncture cells and dispersed by wind. They do not germinate again on barberry plant in any case.
- They **do not germinate again on barbery plant in any case.**
- They infect only primary host i.e. wheat plant.



Life cycle of *Puccinia graminis*

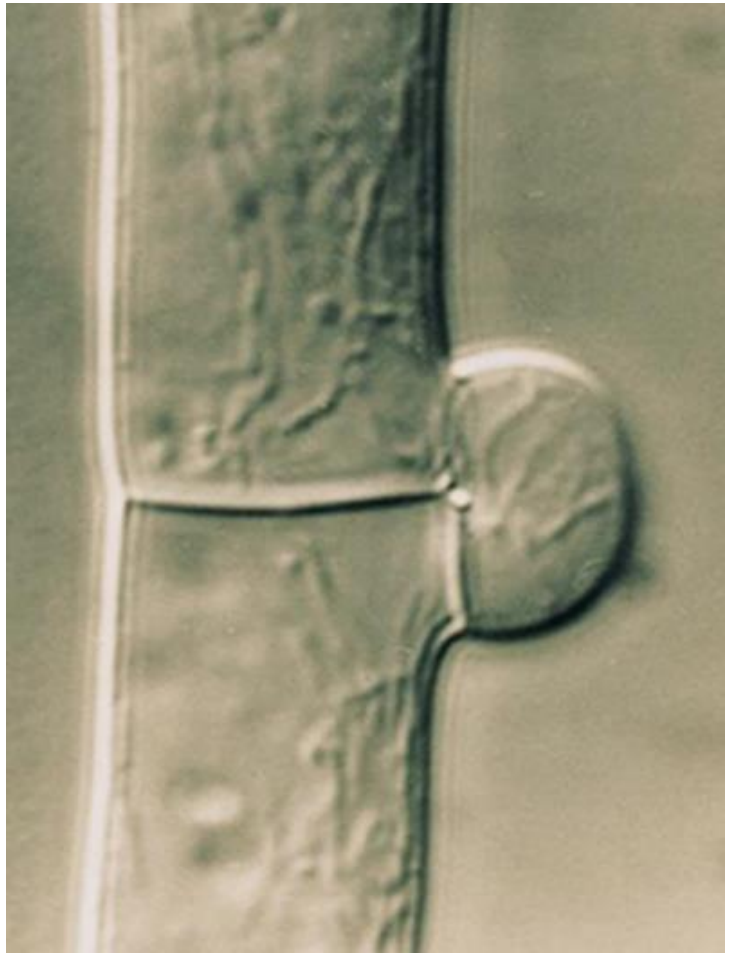
Life cycle

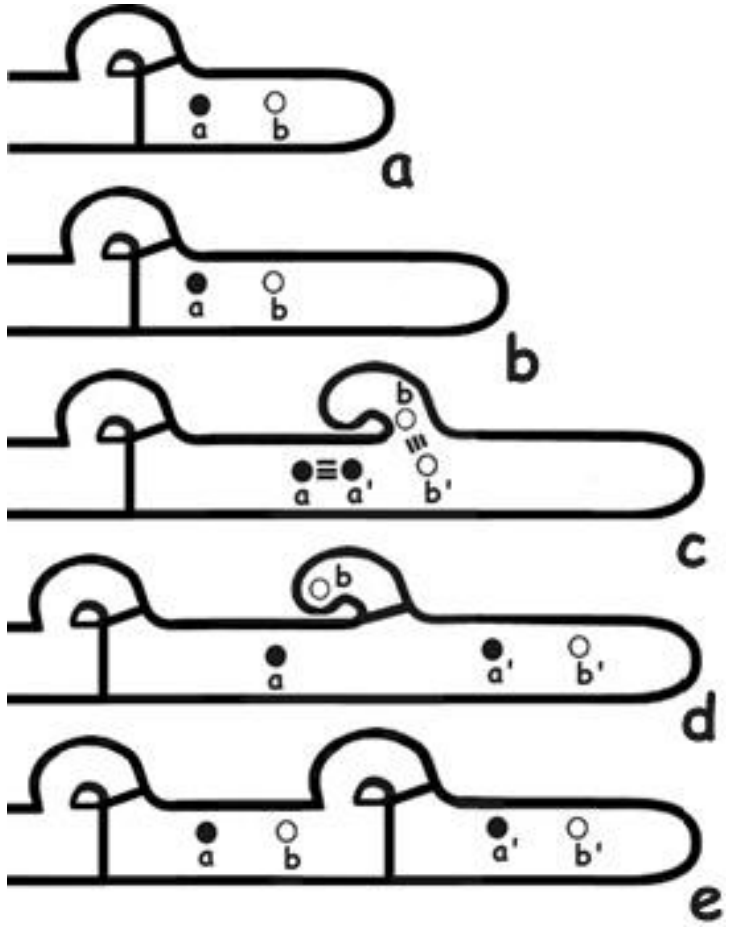

Germination on barberry leaves
Basidium
Basidiospores
Mycelium
(+ and - strain)
Spermogonium
(-)
spermogonium
(+)
Meiosis
HAPLOPHASE
Germination
Recpetive
Hyphae
(-)
spermatium
(+)
Receptive
Hyphae
(-)
spermatium
(+)
spermatization
Mature uninucleate
teleutospores
Karyogamy
Dikaryophase
Dikaryophase
Mycelium
(Dikaryotic)
Dissemination
Rain or wind
Uredospores
Germination
Aecial cup
Bicelled
Teleutospores
Uredosorus
stomatal penetration
Aeciospores
Teleutosorus
Mycelium
(Dikaryotic)
Infection on wheat plant
(Primary host)
germination
dissemination by wind or Rain

Important terms

- **Host:** An Organism which harbors parasite or pathogen and from which it **obtains its nutrition.**
- **Primary host:** The host plant which is **economically** important and **on which Uredospore and teleutospores are produced.**
- **Alternate host/ secondary host**: The other host in the life cycle on which **spermagonia and aecia are produced**
- **Alternative host:** The **host that a pathogen can infect in place of the primary or alternate hosts**
- **Heteroecious** : Organisms with primary and alternate host
- **Autoecious:** Organisms that have **only a single (primary) host**
- **Macrocyclic rust**: **long cycle rust. Produce 5 spores types**
- **Demicyclic rust**: Medium cycle rust. **Omits Uredia**
- **Microcyclic rust**: short cycle rusts. Produce **basidiospores, teliospores and spermatia**

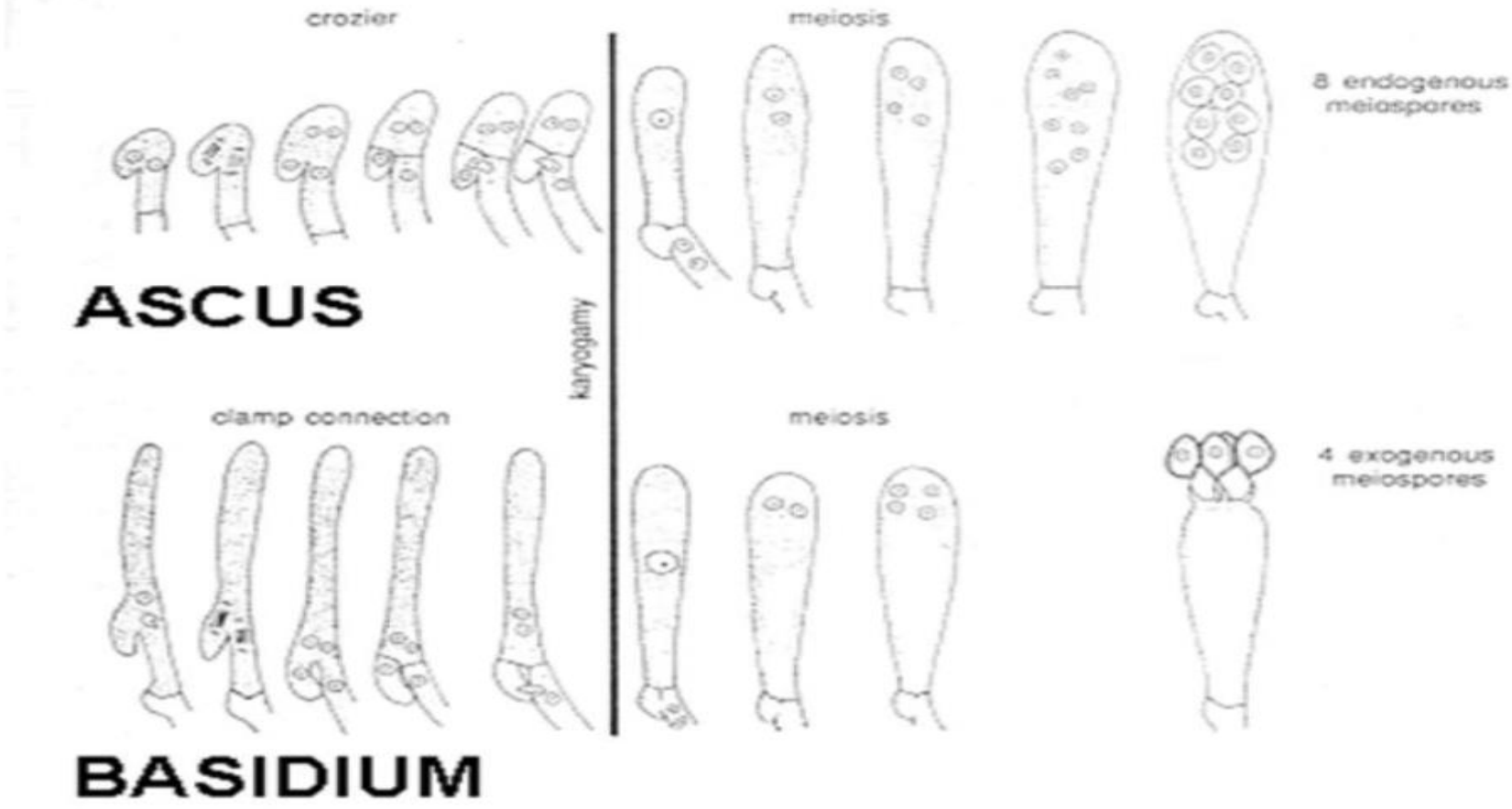
Clamp Connection



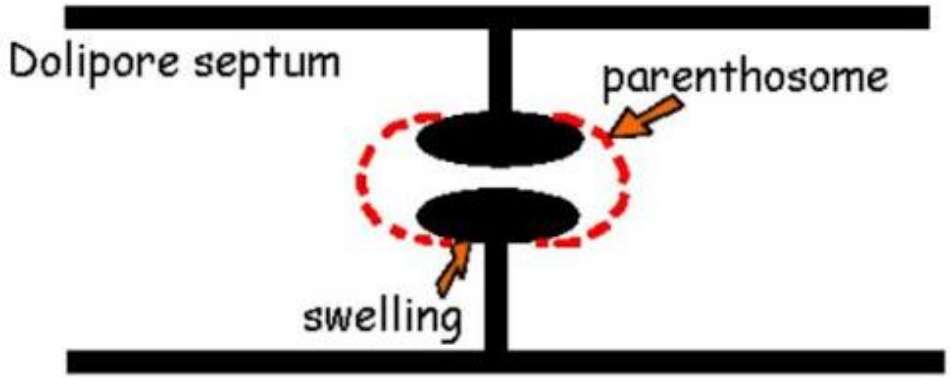


- A **clamp connection is a hook-like structure formed by growing hyphal cells of certain fungi of Basidiomycetes**.
- It is created to ensure each cell, or segment of hypha separated by septa (cross walls), receives a set of differing nuclei, which are obtained through mating of hyphae of differing sexual types.

Difference between Crozier and Clamp connection



Dolipore septum



- Dolipore septa are **specialized dividing walls** between cells (septa) found in almost all species of fungi in the class **Basidiomycetes**.
- Unlike most fungal septa, they **have a barrel-shaped swelling around their central pore, which is about 0.1–0.2 µm wide.**

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Thank You