

## **LECTURE NO.1**

### **MYCOLOGY**

The term "Mycology" etimologically means "the study of fungi". Fungi are highly evolved forms of microorganisms included in the division "Thallophyta" in the botanical classification of the plant kingdom. It is interesting to note that many of the botanist have specialised in mycology and plant pathology. The first Indian universities that were established in 1857 at Calcutta, Bombay and Madras emphasised fungal taxonomy. The organized teaching in mycology and plant pathology as a part of agricultural sciences was being undertaken by the Indian Agricultural Research Institute. Fungi are ubiquitous in nature i.e. they are present in soil, water, air etc

### **PLANT PATHOLOGY**

The term 'Pathology' is derived from two greek words 'pathos' and 'logos'. 'Pathos' means suffering and 'logos' means the study / to speak / discourse. Therefore if etymologically means "study of suffering". Thus the plant pathology is the "study of suffering plants". When the plant is suffering i.e. not developing and functioning in the manner it is expected, then it is called as diseased. Due to this abnormality, the productivity of the plant is reduced or lost.

Plant Pathology (or) Phytopathology is one among the branches of agricultural science that deals with cause, etiology, resulting losses and management of plant diseases with four major objectives.

1. Study the disease(s) / disorders caused by biotic and abiotic agent(s)
2. Study the mechanism(s) of disease development
3. Study the interaction between plant and the pathogen in relation to the overall environment
4. Develop suitable management strategy to surmount the diseases and to reduce the loss.

5. Nature and concept of disease is very much varied. Many scientists proposed a number of definitions for a diseased plant. Anon (1950) defined the disease as "harmful deviation from normal functioning of physiological processes". According to Stakmann and Harrar (1957) plant disease is "a physiological disorder (or) structural abnormality that is deleterious to the plant / to any of its parts / products / that reduces their economic value".

In general disease is an interaction among the host, parasite and the environment.

The earliest known writings make references in the ravages of plant diseases. There are references in the religious literature about the occurrence of plant diseases. Rigveda, Atharveda (1500-500 BC), the Artha Shastra of Kautilya (321 -186 BC), Vishnu Puran (500 AD), Agnipuran (500-700 AD) etc are some ancient books in which they have mentioned the plant diseases and other enemies of plants along with the methods to control them. The man in the Vedic period were aware of the fact that these diseases are caused by microorganisms. Symptoms of plant diseases are also mentioned in Holy Bible, Shakespear's poems and drama's. The early man considered the disease to be the curse of God on man for his wicked deeds. The early man celebrated "Robigalia", a special holiday during which they offered scarifies of red dogs and sheep in an attempt to please the rust god "Robigo" and keep him from sending the rust disease to their crop.

Theophrastus, who lived from 370 BC to about 286 BC was the first scientist to study and write about diseases of trees and cereals. He observed that certain plant species are more liable to certain diseases than others. From the 5<sup>th</sup> century A.D until the Renaissance, there was lack of intellectual activity and little was written on plant pathological aspects. **Ibn-al-aivam** described the symptoms of many diseases of trees and vines during 10<sup>th</sup> century AD. The period between 300 AD and 1300 AD is said to be the middle / dark ages in the history.

After the invention of the compound microscope in the mid-1600's which enabled scientists to see many of the previously invisible microorganisms Scientists as well as lay people became even stronger believers in spontaneous generation of diseases and of

the microorganisms associated with diseased or decaying plant, human or animal tissues. That is, they came to believe that the mildews, rusts, decay, or other symptoms observed on diseased plants, and any microorganisms found on or in diseased plant parts, were the natural products of diseases that just happened rather than the cause and effect of the disease. Late blight of potato (*Phytophthora infestans*) caused severe potato losses in much of Northern Europe in the 1840's but it absolutely destroyed the potato crop in Ireland in 1845 and 1846 and caused the great "**Irish famine**". The destruction of potatoes caused widespread famine that resulted in the death of hundreds of thousands of people and the emigration of more than one and half million people from Ireland to the United States. Wheat rust has been another disease that has appeared in epiphytic form from time to time in many countries. In the fag end of the second world war (1943), Bengal had to face a serious famine (**Bengal famine**). One of the reasons to which this famine has been attributed is the loss in the yield of rice due to attack of *Helminthosporium oryzae* leaf spot disease which had been affecting the crop for last several years. In 1867 coffee rust attacked the plantations in ceylon. The country lost crores of rupees annually and the farmers were advised to take up tea planting. Tea was also attacked by blister blight but by that time chemical control measures were known. In India the current important diseases which are responsible for yield loss are rust and smut in wheat, blast and bacterial blight of rice, downy mildew of sunflower, red rot, wilt, grassy shoot and ratoon stunting disease of sugarcane, virus diseases of pulses, potatoes, banana etc.,

These instances of plant disease epidemics are worth mentioning because they paved the way to strengthen and widen the research on Plant Pathology. During the present century interest in Plant Pathology has also increased dramatically. Now, the science Plant Pathology is branched as Mycology, Plant Bacteriology, Plant Virology, Forest Pathology, Plant Nematology etc., This science is also related to most of the old and new branches of science. By studying the history of Plant Pathology one can get a better perspective of the subject, and came to know to the contribution made in this field and problems that were encountered. For easy understanding, hisotrical

developments in the various fields of Plant Pathology is discussed in different titles here under.

### **Mycology**

- 1675 - Dutch worker LEEUWENHOEK developed the first microscope
- 1983 - LEEUWENHOEK described bacteria
- 1729 - Italian botanist MICHELI studied fungi and saw their spores and also called as founder of mycology.
- 1755 - French botanist TILLET published a paper on bunt or stinking smut of wheat
- 1807 - French scientist PREVOST proved that disease are caused by microorganisms
- 1801 - PARSOON They engaged in the classification and
- 1821 - FRIES nomenclature of fungi and  
discovered the life cycle of bunt fungus
- 1842 - VONMARTIUS They believed that late blight of potato was caused by the
- 1845 - MORREN fungus
- 1846 - BERKELEY
- 1866 - ANTON DE BARY (Germany) was the first to indicate the nature of obligate and facultative forms. He laid the foundation of modern experimental plant pathology. He studied about the blight of potato, heteroecious nature of rust fungi, downy mildew diseases. He trained.  
M.S. WORONIN of Russia, O.BREFEILD of Germany, A. MILLARDET of France, H.M. WARD of England, W.G. FARLOW of the USA and FISCHER of Switzerland. He is truly regarded as the "FOUNDER OF MODERN MYCOLOGY".
- 1875-1912 BREFEILD discovered the methods of artificial culture of microorganisms.
- 1878 - Downy mildew of grape vines was introduced into Europe from America.  
The disease almost ruined the wine industry.

- 1880 - H.M. WARD emphasized the role of environment in the epidemiology of coffee rust. He may be reckoned as he "FATHER OF TROPICAL PLANT PATHOLOGY".
- 1874 - ELIAS FRIES - Linnaeus of mycology.
- 1882 - ROBERT HARTIG contributed on many aspects to forestry, botany. He is aptly called as "FATHER OF FOREST PATHOLOGY".
- 1885 - PIERRE MARIE ALEXIS MILLARDET accidentally discovered the Bordeaux mixture for the control of downy mildew of grapes.
- 1894 - Swedish scientist ERIKSON described the phenomenon of physiologic races.
- 1900 - ORTON                    They worked on control of rusts and wilt diseases of
- 1905 - BIFFEN                    cotton. They are pioneers in genetics of plant disease resistance.
- 1912 - BURGEFF stated that within a cell of the fungus fusion between dissimilar nuclei can occur. This phenomenon is called "heterokaryosis".
- 1914 - E.C.STAKMAN - Contributed valuable information on physiological races of pathogen
- 1920 - E.J.BUTLER initiated an exhaustive study on Indian fungi and the diseases caused by them. He can also be called as the "FATHER OF MODERN PLANT PATHOLOGY IN INDIA". He is the first director of Imperial Mycological Institute (now International Mycological Institute) in England.
- 1923 - HANSEN and SMITH - demonstrated the origin of physiologic races through heterokaryosis.
- 1934 - W.H. TISDALE and I. WILLIAMS revolutionized the study of organic fungicides by discovering alkyldithiocarbamates.
- 1943 - DIMOND, HEUBERGER and HORSFALL discovered the alkylene bisdithio carbamates.
- 1955 - FLOR contributed on gene to gene hypothesis of diseases resistance and susceptibility.

1948 - B. B. MUNDKAR started Indian Phytopathological Society with its journal Indian Phytopathology.

1956 - J. G. HORSFALL published a book entited "Principles of Fungicidal action"

1970 - S.D. GARETT investigated in the management of root diseases and he contributed in the field of biological control.

1886-1971 J.F.DASTUR is the first Indian Plant Pathologist who is credited with detailed study of fungi and plant diseases. His special field of study was the genus *Phytophthora*. Other important contributions.

1953 - J.C. LUTHRA - developed solar heat treatment for wheat loose smut.

### **Plant Bacteriology**

1876 - LOUIS PASTEUR and ROBERT KOCH - They proved that anthrax disease of cattle was acused by specific bacterium.

1876 - ROBERT KOCH of Germany described the theory called "KOCH'S POSTULATES." He established the principles of pure culture technique.

1882 - Americal Plant Pathologist - T. J. BURRUILL first time proved that fire blight of apple and pear was caused by a bacterium (now known as *Erwinia amylovora*)

1905-1920 E.F.SMITH of U.S.A gave the final proof of the fact that bacteria could be incitants of plant diseases. He also worked on the bacterial wilt of cucurbits and crown gall disease. He is also called as "Father of Phytobacteriology".

Chilton and his coworkers demonstrated that crown gall bacterium transforms plant clel to tumour cell by introducing into them a plasmid.

### **Plant Virology**

1886 - ADOLF MAYER - He born in Germany and worked in Holland on Tobacco Mosaic Virus of tobacco. He demonstrated the sap transmission of the disease.

1892 - DIMITRII IVANOWSKI (Russia) demonstrated that the agent of tobacco mosaic virus could pass through even those filters that retained bacterial cells.

- 1895 - E.F. SMITH of U.S.A showed the peach yellows was a contagious disease.
- 1898 - BEIJERINCK - a Dutch microbiologist and founder of virology proved that the virus inciting tobacco mosaic is not a microorganism. He believed it to be *contagium vivum fluidum* (infectious living fluid)
- 1935 - W.H.STANLEY proved that viruses can be made as crystals. He got Nobel Prize.
- 1936 - BAWDEN F.E. and Pirie (Britain) found that the crystalline nature of the virus contains nucleic acid and protein.
- 1939 - Kausche and colleagues first time saw the TMV virus particles with the help of microscope.
- 1956 - GIERER and SCHRAMM proved that the nucleic acid fraction of the virus is actually the infectious agent.
- 1959 - MUNDAY succeeded in inducing TMV mutations.
- 1966 - KASSANIS discovered the satellite viruses.
- 1967 - DIENER and RAYMER discovered the potato spindle tuber was caused by small naked SSRNA which he called viroid.

### **Phytoplasms**

- 1898 - NOCARD and ROUX discovered another agent of diseases (Mycoplasma Like Organism) caused in animals which was similar to viruses in size but which could be cultured on artificial media. This group was known as mycoplasma.
- 1967 - Doi and Ishie, the Japanese scientists found that mycoplasma like organisms (MLO) could be responsible for the disease of the yellows type. Doi observed that MLO's are constantly present in phloem while Ishie observed MLO's temporarily disappeared when the plants are treated with tetracycline antibodies.

### **Spiroplasma**

Davies *et al.*, 1972 observed that a motile, helical wallless microorganism associated with corn stunt diseases, which could be cultured and characterised and they named it as spiroplasma.

### **Plant nematology**

- 1973 - Needham (England), first time reported the wheat gall disease is associated with nematode (now called as *Anguina tritici*)
- 1857 - C. Devaine – studied the life cycle of *A. tritici*
- 1892 - Atkinson reported the association of root knot nematode with the fusarial wilt of cotton.
- 1901 - Hunger showed that bacterial wilt of tomato was facilitated by root knot nematode
- 1958 - Hewitt *et al.* discovered nematode as vector of plant diseases.

### **CONTRIBUTION OF PLANT PATHOLOGISTS IN INDIA**

- |  |   |
|--|---|
| J.F. Dator<br>N. Prasad                    | - Late blight of potato                                   |
| G.S. Kulkarni<br>D. Suryanarayana          | - Downy mildew of sorghum                                 |
| B.B. Mundkur<br>C.V. Subramanian           | - Wilt of cotton – (Book) <b>Fungi and Plant Diseases</b> |
| K.C. Mehta                                 | - Wheat Rusts   |
| J.C. Luthra                                | - Solar seed treatment of wheat seeds                     |
| S.Y. Padmanabhan and<br>S.B. Chattopadhyay | - Diseases of rice  |
| M.J. Thirumalachari                        | - Antibiotics in plant disease - control.                 |
| R.N. Tandon                                | - Diseases of fruits and vegetables                       |
| M.K. Patel<br>M.K. Hingorani               | - Bacterial diseases of crop plants                       |

- P.N. Patel  
G.S. Kulkarni
  - Bacterial diseases of crop plants
- P.R. Mehta
  - Diseases of cereals and millets.
- H.K. Saksena
  - Gram rust and Rhizoctonia diseases
- D.N. Srivastava
  - Bacterial blight of paddy
- R.S. Singh
  - Pythium - Plant Diseases – (Book).
- Y.L. Nene
  - Khaira disease, Downy mildew
  - " Fungicides in Plant Disease control" – (Book)
- V.V. Chenulu
- S.P. Raychavdhuri
  - Virus diseases
- Kapoor and A. Varma
- S.B. Mothur
  - Seed pathology
- G. Rangaswami
  - Diseases of crop plants in India (Book)
  - Bacterial Plant Diseases in India (Book)
- R.K. Saksena
  - Fungal root diseases
- A. Mahadevan
  - Biochemical changes in diseased plants and enzymes.
  - "Growth Regulators, Microorganisms and Diseased Plants" (Book)
- Tisdale
  - Dithiocarpamate - Thiram
- Horsfall
  - Fungicides and Their action (Book)
  - Principles of fungicidal action (Book)
  - Plant Pathology - An advanced Treatise (Book)
- Stanley
  - Purification and Crystallization of TMV.
- Walker
  - Onion - "Plant Pathology", Vegetable diseases (Book)
- Gottlieb
  - Antibiotics.
- Muller
  - Hyper sensitivity and phytoalexins.
- Vander Plank
  - Host resistance
- K.Maramorosch
  - Virus transmission
- Davis *et al.*,
  - Spiroplasma
- E.J. Butler
  - "Fungi and Diseases in Plants"

## MILESTONES IN THE HISTORY OF PLANT PATHOLOGY

### **Mycology**

- 1773 - **Zallinger** gave the systematic classification of plant diseases
- 1807 - **Prevost**, first use copper sulphate treatment for grain smut
- 1821 - **Robertson**, first identified the use of sulphur as fungicide
- 1840 - Tulasine brothers (Louis Rene Tulasne and Charles Tulasne), French botanist first made illustrated drawings of rust, smut and ascomycetes. They have also confirmed the work of prevost on wheat bunt fungus. Among this L.R. Tulasne is called as reconstructor of mycology.
- 1858 - **Julius Gotthelf Kuhn** : Published first text book plant pathology namely "The diseases of cultivated crops. Their causes and their control".
- 1869 - **Raulin** discovered the first synthetic medium for culturing microorganism.
- 1876 - **Farlow**, first introduced independent course of plant pathology at Harward University.
- 1880 - **Frau Hesse** introduced the use of agar in microbiological methods.
- 1887 - **Jensen** developed hot water treatment for smut disease
- 1909 - A.H.R. Buller studied the physiology and fungi.
- 1951 - **Hatson tentter** pioneers to cultivate cedar apple rust fungi on artificial medium.
- 1956 - **Muller** coined the term Phytoalexin for the post infectional chemicals that have significant role in disease resistance.
- 1962 - G.Farkars and Z. Keralay identified the role of phenolic compounds in natural chemical resistance in plants.

### **Bacteriology**

- 1876 - **Robert Koch and Pasteur** disproved the theory of spondaneous generation of diseases and propose germ theory in relation to the diseases of man and animal.

- 1876 - **Robert Koch;** German physician established principle of pure culture technique, plate method of isolation of bacteria and fungi and first man to culture a bacterium capable of souring milk. he described the theory called koch postulate.
- 1882 - **T.J. Butil** first teach the plant pathology at the university of *illinois*
- 1891 - **Waite** discovered transmission of virology, *Erwinia anglovera* (fire blight of apple, plum, pear) by bees and wasps.
- 1902 - Takami discovered the insect transmission of rice stunt virus.
- 1936 - **Kunkel** from USA used hot air treatment to eliminate plant viruses.
- 1955 - **Homes**, obtain virus free stock by tip culture method.
- 1966 - **Kassanis**, pioneer in apical meristem culture to eliminate viruses.

### **Nematology**

- 1959 - Raski, introduced nematological studies as a part of plant pathology.

### **Virology**

- 1915 - F.W. Twort – discovered the first disease of bacteria.
- 1917 - **d'Herelle** coined the term bacteriophage
- 1976 - Shepherd, R.J. – studied the DNA viruses of many higher plants.

## **LECTURE NO.2**

### **Classification of Plant disease**

On the basis of causes of plant diseases mentioned above they can be classified into following two categories.

#### **1. Non-infectious diseases**

These are diseases with which no animate or virus pathogen is associated. Therefore they remain non-infectious and cannot be transmitted from one diseased plant to another healthy plant. Since no parasite is associated with these diseases they are also known as non-parasitic diseases.

#### **Examples**

Tip rot or Necrosis of mango due to boron deficiency.

Black heart of potato due to unfavourable oxygen relations in stores and in field.

Khaira disease of paddy due to non-availability of zinc to the plant.

## **2. Infectious diseases**

These are diseases, which are incited by foreign organisms under suitable environments. A specific pathogen is responsible for the diseases. These diseases are always infectious, sometimes contagious and are transmitted from diseased to healthy plants in the field and from one place to another through various agencies. When an animate cause is isolated or identified, its pathogenicity must be proved to confirm that it is the actual cause of the disease. There are many chances that more than one organism will be found in or on the diseased organ. Robert Koch (1882), a German scientist proposed a set of rules to demonstrate the association of a microbe with the disease in a host. These rules, known as Koch's postulates.

### **Koch's postulates are:**

1. The pathogen must show constant association with the disease.
2. The pathogen must be isolated from the diseased host and grown in pure culture.
3. The pathogen from the artificial culture should be able to reproduce the disease when inoculated on healthy plant of the same kind. The symptoms produced should be identical with those seen on the plant from which isolation was made.
4. The artificially produced disease should yield the same pathogen on reisolation.

Infectious diseases are often classified according to their occurrence in the following groups.

### **Fungi**

Fungi are eukaryotic spore bearing achlorophyllous organisms that may reproduce sexually and asexually and whose filamentous branched somatic structures are typically surrounded by cell walls containing chitin cellulose or both of these substances with many other complex carbohydrates

## BACTERIA

Bacteria are primitive organism classified as prokaryotes and their nuclear material is not separated from the cytoplasm by a nuclear membrane and there is no mitoticx apparatus.

The prokaryotes differ from eukaryotes in many ways as shown in the Table.

Sl.No.	Prokaryotes	Eukaryotes
1.	Show primitive cellular organisation	Possess advanced cellular organisation
2.	Cells small sized 1 to 2 x 1 to 5 micron or less (1 micron = 1/000 mm)	Cells are larger than prokaryotes (greater than 5 micron in width or diameter)
3.	Cell wall is made up of peptidoglycan (mycopeptide).	Cell wall is made up of cellulose
4.	Membrane bound organelles such as endoplasmic reticulum, golgi complex, mitochondria, chloroplasts and vacuoles are absent.	Membrane bound organelles such as endoplasmic reticulum, golgi complex, mitochondria, chloroplasts and vacuoles are present
5.	Ribosomes are smaller and made of 70S units (S refers to Svedberg unit, the sedimentation coefficient of a particle in the ultra centrifuge)	Ribosomes are larger and made of 80S units. But ribosomes of mitochondria and chloroplasts are of 70S type.
6.	Do not possess an organised nucleus. Nuclear membrane and nucleolus are absent	Possess an organised nucleus. Nuclear membrane and nucleolus are present
7.	Genetic material (DNA) is not found in well organised chromosomes	Genetic material is found in well organised chromosomes
8.	Deoxyribo nucleic acid (DNA) is shorter and circular. Not histone (protein) bound	DNA is long and linear. Histone, bound
9.	Cells divide only by fission (amitosis).	Cells divide by mitosis and meiosis
10.	Flagellum is of single fibrillar type	Flagellum is multifibrilled containing eleven (9+2) pairs of microtubules
11.	Vegetative and asexual reproduction are most common. Sexual reproduction is usually absent. Only transfer of genetic material by conjugation occurs in certain bacteria	Besides vegetative and asexual reproduction, sexual reproduction also occurs.

## **VIRUS**

A virus is a nucleoprotein that is very small to be seen with a light microscope multiply only in living cells and has the ability to cause disease.

## **VIROIDS**

Viroids are small low molecular weight ribonucleic acids that can infect plant cells replicate themselves and cause disease.

## **PHYTOPLASMA**

They lack cell wall, are bounded by a unit membrane and have cytoplasm ribosomes and strands of nuclear material. They are pleiomorphic.

### **Endemic disease**

The word "endemic" means "prevalent in, and confined to, a particular country or district" and is applied to disease. A disease is classified as endemic when it is constantly present in a moderate to severe form and is confined to a particular country or district.

E.g. Club root disease of cabbage is endemic in the Nilgiris district.

### **Epidemic or Epiphytotic disease**

A disease usually occurs widely but periodically in a destructive form is referred as epidemic or Epiphytotic disease.

E.g. Powdery mildew disease in grapevine.

### **Sporadic disease**

Sporadic disease is one which occur at very irregular intervals and locations and in relatively fewer instances. In reality, sporadic disease belongs to the epidemic group.]

E.g. Udubatta disease in rice.

### **Pandemic disease**

These occur all over the world and result in mass mortality. '

E.g. Damping off disease in Tomato.

## LECTURE NO.3

### **Fastidious vascular bacteria**

These are previously called as rickettsia like organism or RLO that cause plant diseases and can not be grown on artificial media in the absence of host cell. Almost off of them are limited only fastidious to xylem/phloem.

### **Phloem inhabiting fastidious bacteria**

Phloem limited bacteria was first observed in 1970 in the phloem of clover and periwinkle affected with clover club disease by *D. lefleche* and J.M. Bowe and in later citrus plants affected with greenurg disease. They produce characteristic symptom like yellows type which include stunting, yellowing of young leaves, virescence of floral parts, premature flowering and fruit drop, witches broom and premature death of the entire plant. They are often transmitted by leaf hoppers, dodder and by grafting but citrus greenurg is transmitted by the citrus *psylla* in addition to vegetative propagation.

This bacteria cells are present mostly in mature sieve elements, irregularly distributed among the vascular bundles. They are mostly rigid rods, non motile, 0.2 - 0.5 x 1.2  $\mu\text{m}$  in size, bounded by double membrane or a cell wall and cytoplasmic membrane. Both membranes are triple layered and are separated by an electron lucent zone. Peptidoglycan is absent and are mostly gram negative.

### **Xylem inhabiting fastidious bacteria**

In 1973, fastidious xylem inhabiting bacteria was observed in the xylem vessels of grape plant affected with Pierces disease and alfalfa affected with alfalfa dwarf. Few other examples are the phony peach disease, sugarcane ratoon stunting, almond leaf scorch etc.

All the xylem inhabiting fastidious bacteria known so far are gram negative but one exception that the sugarcane ratoon stunting is a gram positive bacteria, and are classified as the members of the genus *clavibacter* (formerly called as

*Corynebacterium*). These fastideous xylem inhabiting bacteria are differentiated from other xylem inhabiting bacteria (*Pseudomonas*, *Erwinia* etc.) by their inability to infect tissues other than xylem, their insect transmission but not mechanical transmission and their inability to grow on bacteriological culture media.

Xylem inhabiting fastideous bacteria are generally rod shaped  $0.2 - 0.5 \times 1 - \mu\text{m}$  in size. The cells usually have a well defined cell wall and plasma membrane both triple layered in structure. The walls are rigid (or) rippled due to periodic unfolding of outer membrane of cell wall. The cell wall consist of an outer membrane an intermediate electron lucent zone and an inner dense peptidoglycan layer which is separated from the plasma membrane by a electron lucent zone. These fastideous bacterial cell also multiply by fussion the transmission of these bacteria is almost entirely by xylem feeding insects exclusively to leaf hoppers such as sharpshooter leaf hopper and spittle bugs. The bacterium is transmitted by the vector in a non circulative but persistent manner. The symptoms of fastideous vascular bacte consist of marginal chlorosis and necrosis of leaves, stunting, general decline and reduced yields. Such symptoms are probably caused by plugging of xylem vessels by the bacterial cells and by a matrix material partly of bacterial and partly of plant origin.

### **Important fastidious vascular bacterial diseases**

#### **1) Xylem inhabiting fastidious bacteri**

##### **i) Gramme positive**

- a) Sugarcane ratoon stunting (*Clavibacter xyle* subsp. *Xyli*).

##### **ii) Gram Negative**

- a) Pierce's disease of grapes

- b) Citrus variegation

These are all caused by the forms of bacterium

- c) Peach phony disease

*xylella fastideosa*

- d) Plum leaf scald

e) almond leaf scorch

## 2) Phloem inhabiting fastidious bacteria

- a) Citrus greening
- b) Clover club leaf (Gram negative)

### **PHYTOPLASMAS, SPIROPLASMAS L – FORM BACTERIA**

For about 90 years after the discovery of first phytopathogenic bacteria some more prokaryotes including mycoplasmas, phytoplasmas, spiroplasmas and L-form bacteria which were in size between bacteria and virus were identified as causal agent of plant diseases. These organisms exist in nature and differ from the true bacteria I the absence of typical cell wall. But their cells are enclosed by unit cell membrane. They are generally called as mollicutes, come under the division *Tenericutes* of the kingdom *Protista*. The division tenericutes contain one order namely *Mycoplasmatales* that has three families viz. *Mycoplasmataceae* *Acholephasmataceae* and *spiroplasmataceae*. Each family has one genus respectively *Mycoplasmas*, *Acholeplasmas* and *Spiroplasmas*.

#### **Mycoplasmas**

Mycoplasmas were known to the field of medical science in 1898 only after the work of Nocard and Roux, who succeeded in culturing the contagious bovine pleuropneumonia organism in artificial media. These organism was later known as *Mycoplasma mycoides* kept under the order *mycoplasmatales* among the bacterial class *Schizomycetes*. Later, the order *mycoplasmatales* was placed in the class *tenericutes* of mollicutes.

The distinguished characteristics of mycoplasmas are.

- Unicellular, gram positive, non motile, highly pleomorphic facultative saprophytes and their size ranges from small, spherical bodies to large irregular bodies (with diameter ranges from 175 – 250 nm. Some species produces branched myceliod structure.
- Free living, both parasitic and saprophytic, and reproduces by budding and binary fission.

- Cell contains cytoplasm, ribosome and nucleic acid devoid of nuclear membrane. Both DN and RNA are present.
- Cell wall is absent but the cells are delineated by a unit lipoprotein membrane of about 10 nm thickness. About 10 amino acids are present in the protein fraction while lipid of all mycoplasms contains sterol, carotenols, glycerophospholipid, glycolipids etc.
- Cells are filterable through bacterial filters, can be cultured artificially on cell free agar medium with sterol and form "Poached egg" or "fried egg" shape colonies with central nipple.
- Usually resistant to antibiotics like penicillin, cephaloridini etc. which acts on cell wall but they are sensitive to tetracycline.
- Reduce tetrazolium chloride in aerobic and or anaerobic culture media.

Regarding plant science, the mycoplasms were identified as plant pathogen during 1967. Prior to 1967, plants showing witches broom phenomenon are considered as viral diseases and that type of diseases are generally called as 'yellows'. Since no fungi, bacteria, protozoa and nematode are found to be associated with such diseases. Moreover their causative agents are transmitted through insect vectors, vegetative propagative material, grafting, parasitic plants etc. and they pass through the bacterial filter of 450 nm diameter. However, all attempts made to purify the organisms by routine virological methods and to characterize them morphologically and chemically as viruses remained unsuccessful.

In 1987, Doi *et al.* and Ishiiie *et al.* from Japan first time identified a well less microorganism in the phloem of mulberry plants infected mulberry dwarf (a kind of yellow disease). They proposed that these organism may be mycoplasma like organism (MLO) or chlamydia like organism because of their superficial resemblance to mycoplasms and therapeutic effect of tetracycline antibiotics in such prokaryotes. When compare to animal mycoplasmas, the mycoplasmas observed in plants have not been cultured. Hence the plant mycoplasmas have not been characterised and koch's postulate have not been proved. Further detected studies revealed that the mycoplasmas

present in plant sieve elements are not true mycoplasmas and now they are differentiated as phytoplasmas (wall less, round to elongate prokaryotes) and spiroplasmas (wall less, motile, helical prokaryotes).

### **Phytoplasmas**

As characterised earlier, they are wall less round to elongate prokaryotes exist in nature, affecting plants and present in phloem vessels. They are toxonomically more related to acholeplasma than mycoplasma. In plants, more than 200 phytoplasmal diseases are recorded till date of which some are very destructive and may lead to cent per cent failure of crop.

### **Characteristics of phytoplasma**

1. They lack cell wall out the cells are bounded by single triple layered highly elastic unit membrane.
2. Have cytoplasm, ribosome (RNA) and strands of nucleic acid (DNA) similar to bacteria.
3. Usually spheroidal to ovoid (or) irregularly tubular to filamentous in shape and their sizes are comparable to true mycoplasmas.
4. Generally present in the sap of a small number of phloem sieve tubes and cannot be grown on artificial nutrient media.
5. Sensitive to antibiotics particularly those of tetracycline group similar to mycoplasmas. The symptom already present in the plant may disappear or recede whenever tetracycline treatment is given. But the same symptom reappear some after treatment stops.
6. Transmitted through vegetative propagative materials, dodder, grafting/budding and also by insect vectors. The leaf hoppers rank first in transmitting phytoplasmas from plant to plant. Other than this, plant hoppers, psyllids, aphids also transmit phytoplasmas. Within the plants, phytoplasmas move through sieve pores or plasmodesmata. Infected plants become totally freed of mollicutes by heat treatment.

## **LECTURE NO.4**

### **GENERAL ACCOUNT OF FUNGI**

Fungi are the eucaryotic protist, achlorophyllus, nucleated, branched, unicellular or multicellular organisms that may reproduce by the division of vegetative cells, well defined asexual and sexual spores. The body of the fungus is called as 'Thallus'.

#### **Plant Hyphae**

It 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 characterised as septate (with cross wall) or coenocytic (aseptate - without cross wall). Hyphae of most of the fungi measure 5-10  $\mu\text{m}$  across.

#### **Dolipore**

It is a pore in the cross wall/septum through which cytoplasm is in continuity among adjacent cells. The region of the septum around the dolipore is swollen and the swollen rim on either side is capped by a membrane which is called as "parthenosome" or verschluss band or septal pore cap.

#### **Mycelium (Plant Mycelia)**

The hyphal mass or network of hyphae constituting the body (thallus) of the fungus is called as mycelium.

#### **Fungal cell structure**

Fungal cells are typically eucaryotic and have distinguished characteristics than that of bacteria, and algae (Table ).

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

cellulose is either lacking or masked by chitin. Calcium carbonate and other salt may also be deposited upon or within the cell wall.

The living protoplast of the fungal cell is enclosed in a cell membrane called as plasma membrane or plasmalemma. projecting into cytoplasm are called as *Plasmalemmosome*. Like other eucaryotic cells, cytoplasm contains organelles such as nucleus, mitochondria, Golgi apparatus, ribosomes, vacuoles, vesicles, microbodies, endoplasmic reticulum, lysosomes, microtubules .

The fungal nucleus have 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 secretory action of golgi apparatus to the site where these products are utilised.

### **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.

(eg) *Pythium aphanidermatum*

### **Holocarpic**

The thalli 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' (eg) yeast, *Synchytrium* sp.

### **Monokaryotic mycelium or 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.

### **M multinucleate**

The fungal cell contains more than 2 nuclei.

### **Modification of fungal hyphae**

In most fungi, during certain stages of its life cycle the mycelium become organised in to loosely or compactly woven tissues which are distinguishable from the normal hyphae composing a thallus.

#### **1) Plectenchyma : (Gr. plekein =to weave + enhyma = infusion)**

Hyphae may have an altered shape and are closely packed together, easily indistinguishable from true tissues of higher plants. Such pseudo tissue are called as plectenchyma.

The plectenchyma is of 2 types.

1. Prosenchyma
2. Pseudoparenchyma

#### **a) Prosenchyma (Gr. Pros = towards + enhyma = infusion).**

If the altered hyphal elements are visible as hyphae and mostly linear cells lie parallel to one another forming a loose tissue and such tissue is termed as prosenchyma

**b) Pseudoparenchyma (Gr. Pseudo - false + parenchyma - a type of plant tissue)**

In some fungi, modified hyphal elements are oval or isodiametric in shape and closely packed.

**2) Rhizomorph (Gr. rhiza - root + morphe - shape)**

In many root invading or wood destroying fungi, hyphae is aggregated longitudinally and form long root like strands of compact masses. These strands are called as rhizomorph which have a thick hard cortex and a growing tip. These are the organs of mycelial migration and food transport and it provides a great inoculum potential to the invading hyphae in either colonization of a dead substrate or infection of a host. (eg) *Armillariella mellea*.

**3) Sclerotium (Pl. Sclerotia)**

It is a hard, compact resting body, made up of mass of mycelium produced in some fungi after a vigorous active growth. They are generally black or purple outside and greyish white inside. They are irregularly shaped and possess rich amount of food reserve (eg) *Rhizoctonia solani*, *Sclerotium rolfsii*, *Macrophomina phaseolina*. Sclerotia may vary from the size of pinhead to 1 cm in diameter. The fungus *Polyporus* produces the largest size of sclerotium as man's head and size as much as 15 kg.

**1) Myceliogenous**

If the sclerotia germinate directly into mycelium then it is called as myceliogenous (eg) *Sclerotium rolfsii*, *Rhizoctonia solani*.

**2) Carpogenous**

Here in this case, sclerotia germinate and gives rise to spore fruit bearing stalk (eg) *Claviceps fusiformis* and *Sclerotinia sclerotiorum*.

In *C. fusiformis*, the sclerotia germinate and put forth small or long stalked stromata with a globose sporogenous head consist of sexual spore (ascospores) enclosed in peritheciun.

## **Stromata**

Any compact hyphal aggregation in or on which fructification are formed are called as stromata. Acervuli, sporodochia are nothing but fertile stromata bearing sporophores and spore. Some kind of sclerotia also produce stromata during its germination as mentioned in *C. fusiformis*.

## **3. Sporogenous**

Sclerotia on germination gives rise to a spore are called as sporogenous (eg) *Botrytis cinerea*.

## **4) Chlamydospore**

Fungi like *Fusarium* sp produce swollen or thickened resting structures by the rounding up of cells (resting cells) and are termed as *chlamydospores*. They are produced either singly or in chains and may be terminal or intercalary in the hyphae.

The rhizomorph, selerotium and chlamydospores are some of the resting spores of fungi that remain dormant over long period and germinate when the favourable condition appears.

## **5. Gemnae**

They are similar in structure to chlamydospore but they are not thick walled, occur either singly or chains and are generally borne terminally eg *Mucor* sp.

## **6. Appressorium (Pl. Appressoria) (L - apprimere - to press against)**

These are localized swellings of the tip of germ tube or older hyphae that develop in response to contact with the host. In simple these are special structures for attachment in the early stage of infection. From these a minute infection peg usually grows and enters the epidermal cell of the host.

## **Haustoria**

They are nothing but organ for absorption. It is the lateral outgrowth of intercellular or superficial hyphae which will help to absorb food and nutrients from

the host. They are of different shapes and size ranging from knob like structures to simple, lobed, branched, coiled and they are able to penetrate only in the cell wall and not in the plasma membrane.

### **Pseudo selerotia**

These are selerotia like structure composed of funghal hyphae and friable material of the substratum bound together. The mummified fruits parasitised by many fungi are actually pseudosclerotia.

## **LECTURE NO.5**

### **TYPES OF PARASITISM**

Fungi are essentially heterotrophic or dependant on other organisms as they are chlorophyll less in nature. Moreover, they require predigested food to live and therefore they obtain food from either living (*Parasites*) or non living sources (*Saprophytes*).

Based on the source of nutrition, the fungi may be classified as.

#### **1) Parasite**

An organism that lives on or in some other living organism (host) and derive nutrition are called as parasites. The relationship between the parasite and the host is called as parasitism. The parasites are of different types.

##### **i) Ectoparasite**

The parasite may live on the external surface of the host (eg) *Erysiphe polygoni*.

##### **ii) Endoparasite**

The parasite may grow inside the host cell (intracellular) or in between the cells (inter cellular) of the host plant and draw nutrition. (eg) *Leveillula taurica*.

### **Facultative parasites**

They usually live as saprophyte but can attack living tissue under certain condition.i.e., they normally grow as saprophyte but also have the ability to parasitise.

These fungi are having saprophytic phase with a short parasitic phase under suitable condition (eg) *Pythium* sp., *Rhizoctonia* sp.

### **Facultative saprophytes**

They usually live as parasite but can grow saprophytically ie they have long parasitic phase and with a short saprophytic phase in their active life cycle (eg) *Phytophthora* sp. *Venturia inaequalis*, *Spacelotheca* sp.

### **Obligate parasite**

They invariably occur as parasites in nature and cannot be grown in axenic culture media under *in vitro* condition. Some of the so called obligate parasites have been now cultured under laboratory condition as such parasites are called as 'biotrophs' (eg) Downy mildew, Powdery mildew.

### **Hemibiotrophs**

These pathogen attack living tissues and grows in or on them in the same way as that of biotrophs but continue to grow and reproduce after the tissue is dead (eg) leaf spotting fungi.

### **Perthotrophs**

They are so called less specialised parasite which can cause immediate and severe damage to their host tissue. They usually obtain nutrients from cells killed in advance (eg) *Rhizopus*, *Pythium*, *Rhizoctonia*, *Fusarium*. They kill the tissue before actually entering and feeding on them.

### **Necrotrophs**

There are some intermediate parasites which invade and kill host cell (eg) *Phytophthora*, *Claviceps*, *Venturia*.

### **Specialised parasite**

They are the more specialised group of parasites which can cause only slight damage first to the host and the host cells are damaged later at the time of sporulation (eg) smut.

## **Phenomenon of fungal parasitism**

### **Synergism**

It means the ability of two kinds of organisms to grow better or produce greater effect conjointly than either one could alone (eg) *Diplodia nataensis* and *Colletotrichum gleosporioides* together produce much greater effect on citrus bark than either one of the pair alone can produce.

### **Symbiosis**

Some fungi derive nutrition from a living member to which they also provide some kind of benefit in return. This kind of relationship is termed as symbiosis (eg) VAM.

## **MYCORRHIZA**

It is one kind of mutualism between the fungi and roots of higher plants. The name 'mycorrhiza' literally means "fungus root" and it was coined by Frank in 1885. They are quite common in nature and majority of the plant species are known to have some form of mycorrhizal system in their root. ie fungal hyphae like in association with the roots either on the periphery or in the cells of certain layers of the cortex.

There are three types of mycorrhizas.

1. Ectomycorrhiza
2. Endomycorrhiza
3. Ect endo mycorrhiza

### **Ectomycorrhiza**

These mycorrhiza forms an external fungal sheath or mantle round the root. The mantle is of considerable thickness and may be differentiated into layers (*Pseudoparenchymatous layer*) differing in compactness and commonly two in number. The hyphal branches from the sheath grow intercellularly into the cortical parenchyma of the host tissue in the form of network known as "Hartig net" named after the German

botanist "Hartig". Hyphal branches from the sheath also moves to the soil. These kind of mycorrhiza is noticed in most of the forest trees like pine, fir, oak etc. Most of the ectomycorrhizal fungi belong to basidiomycotina and some to ascomycotina (eg) *Amanita* spp, *Boletus* sp, *Pisolithus tinctorius*, *Marasmius scorodonius*.

### **Endomycorrhiza**

These mycorrhiza does not form external sheath or mantle but the hyphae penetrate both between (inter cellular) and with in (intracellular) tissues of cortex and epidermis. The endomycorrhiza may have septate (mostly basidiomycotina) or aseptate mycelium (mostly zygomycotina). Most common aseptate endo mycorrhiza are known as vesicular - arbuscular mycorrhiza (VAM). After germination the aseptate hyphae colonize the cortex, both inter and intracellularly and it branches repeatedly in a dichotomous manner to form a complex branched haustorium called as arbuscules. The hyphae within the cells or intercellular spaces also swell to form thick walled vesicles, that contains fat globules due to the formation of vesicles and arbuscules. These mycorrhiza are named as VAM.

*Endogone* sp. *Gigaspora* sp, *Glomus* sp. *Acaulospora* sp. *Gigaspora margarita*, *Glomus mosseae*, *G. fasciculatus*, *Ectendomycorrhiza*. They have characters of both endo and ectomycorrhiza.

In this case, they usually form a mantle or sheath. The external hyphae penetrate between the epidermal and cortical cell constituting the Hartig net just like ectomycorrhiza and also penetrate into the active living cell similar to endomycorrhiza. (eg) *Coenococcum geophilum*.

In all the three types, the hyphae of the fungal symbionts permeate the soil and obtain scarce and relatively immobile nutrients especially phosphorus but also nitrogen, potassium, Cu, Zn and supplied more effectively to the plants than the root hairs of the concerned plant.

## REPRODUCTION IN FUNGI

Fungi reproduce both asexually (without a nuclear change) and sexually (with a nuclear change). The reproductive bodies are called as spores which are capable of growing into a new thallus. The reproductive phases starts when the vegetative phase (*Somatic phase*) has reached certain degree of maturity so as to provide sufficient quantities of reserve food for the spore producing organs or sporogenous organs. Usually the spore producing organs have.

- ❖ limited growth, vertically oriented and perpendicular to the plane of mycelial growth.
- ❖ Endowed structurally with differentiated cellular characters capable of producing one or more spores.

The reproduction in fungi takes place by the following methods.

- ❖ Asexual reproduction
- ❖ Sexual reproduction

### **II) Asexual reproduction**

It is otherwise called as vegetative or somatic reproduction and it does not involve the union of sex organs (gametangia), sex cells (gametes) or nuclei.

The spores produced by asexual reproduction are called as *mitospores* since they are formed by mere mitotic division of nuclei and the daughter cells have the genetic constituent as that of original cells. In general asexual reproduction occurs many times in the life cycle of the fungus compared with sexual reproduction which occur once in their life cycle. A sexual spores are one or more celled containing one or more nuclei in each cell with same protoplasmic contents. Some fungi produces more than one form of asexual spores and each one develop into thalli similar to mother thalli in all the developmental cycle. Such fungi are called polymorphic or pleiomorphic. The fungi *Fusarium* produces micro and macroconidia as its asexual spore. Spores are

borne on spore bearing structures called as *Sporophore* which is a special branch of vegetative hyphae.

Sporophore may be simple or compound and they bear sprogenous cells. Sporophores either have determinate or indeterminate growth. The asexual spores differ in size, shape, function and mode of formation. The identification and classification of fungi are largely based on the structure of spore and spore bearing hyphae.

In fungi, the asexual reproduction takes place by the following methods.

### **Fission of unicellular thalli**

Some types of yeast undergoes this type of reproduction. Nuclear of a somatic cell divides mitotically and the cell contents become divided into two halves by the formation of transverse septum. Later the two separated cells get round off and become individual thalli.

### **2. Reproduction by budding**

It is also a common method of reproduction in *Saccharomyces* spp. The unicellular thalli put forth one or two buds which enlarge gradually. The protoplasmic contents including mitotically divided nuclei pass into the bud, and then the buds cut off to become a new thalli.

### **3. Fragmentation**

Hyphae break up into their component cells which behave like spores. These spores are called as *arthrospores* or *oidia* (eg). *Oidium* species.

### **4. Production of asexual spores**

Fungi produce different types of asexual spores viz., (Zoospore, *aplanospore* *sporangiospores* and conidia. Out of this, the zoospores and aplanospores are produced endogenously inside the sac like structures (*Sporangia*) while conidia is produced exogenously on a special hyphae called as *conidiophore*. The sac's bearing zoospores are known as zoosporangia and aplanospore are called as sporangia.

## **Sporangium (Pl. Sporangia)**

It is a sac like structure containing spores either aplanospore or zoospore and are usually formed at the end of spore bearing structures called *sporangiophore*. Sporangia are produced either singly (*Pythium* sp, *Phytophthora* sp) or in chains (*Albugo* sp.) and are pigmented (*Rhizopus* sp) or hyaline (*Pythium* sp, *Phytophthora* sps). Sporangiophore may be sympodially branched, (*Plasmopara viticola*), dichotomously branched (*Peronospora parasitica*), or monopodial branched (*Rhizopus* sp) etc.

Spores produced from the sporangia (zoospore/aplanospore) are always one celled uninucleate or multinucleate.

## **Zoospore (Motile spore/Planospore/Swarm spore)**

It is a motile asexual spore produced mostly by the lower fungi belonging to mastigomycotina subdivision. They have one or two flagella for motility in the surrounding film of water and are spherical or reniform or pear shaped.

Zoospores have hyaloplasm membrane cover and are uninucleate and haploid. The flagella may be anterior or posterior (opisthocont) or laterally attached to a groove of the body. Within the body of the zoospore the flagellum is attached to a basal granule called as *blepharoblast*. Two types of flagella are found. 1. *Whiplash* (Acronematic or Peitchgeisal flagella) 2. *Tinsel*. (Pantonematic or flimmer or flimmer geisel flagella)

Whiplash flagellum have a long rigid base with a short flexible end. While the tinsel flagellum is feather like with a central rachis (axoneme) which is covered on all sides along its length with short sub fibrils (flicker hairs or mastigoneme).

In some fungi, zoospores are not produced directly inside the sporangium. Instead of that, the inner wall of the sporangium may grow out into a short or long tube which swells to form a vesicle. The entire sporangial content move into the vesicle,

nucleus divides mitotically and each of the nuclear become a zoospore (eg) *Pythium* sp.

After released from the sporangia or from the vesicle, the zoospore pass through three distinct phases *viz.*, motility, encystment and germination. Whether the zoospore is *monoplanetic* or *diplanetic*, germination is always preceded by encystment which follows a period of active movement of the spore.

*Monoplanetic* means fungi with only one swarming stage involving the formation of one kind of zoospore (eg) *Pythium*. Fungi have two swarming stage with two kind of zoospores, then it is called as *diplanetic*. For example *Saprolegnia* sp. produce two kinds of zoospores in their life cycle. The primary zoospores are pear shaped, secondary one are kidney shaped. Those which have more than 2 swarming stge are called as *Polyplanetic* (eg) *Achlya* sp.

The body of the zoospore after loss of motility, become more or less spherical and secrete a thick wall around itself which is called as *encystment*. In some parasitic fungi, the zoospores are attracted towards the host surface due to some chemical stimulants released by the host tissues and that process is called as *chemotaxis*.

Zoospores aid in initiating new generation and spread of the fungus. But in some lower fungi, they also act as gametes.

### **Apalanospore**

These are non motile asexual spores produced by many type members of zygomycotina fungi inside the sporangia. They are either uninucleate or multinucleate, formed only at the periphery of the sporangium with a empty centre and base which is called as *columella* (eg) *Mucor* sp, *Rhizopus* sp.

### **Conidia**

Conidia are the asexual spores produced by most of the higher fungi (*terrestrial fungi*) including ascomycotina, basidiomycotina and deuteromycotina. But in

basidiomycotina, the asexual spore production is rare. Generally the term conidia is used for any asexual spore other than sporangia and spores formed directly by the hyphal cells.

Conidia are formed singly or in chains (acropetal succession/basipetal succession) on a special spore bearing hyphae called as *conidiophore*.

In basipetal succession, the oldest conidia is present at the apex of the conidial chain (eg) *Erysiphace* sp. On contrary, oldest conidia is present at the base in case of acropetal succession (eg) *Cladosporium* sp. Conidia borne as buds or blown out tips in acropetal chains are also called as *blastospore* (eg) *Cladosporium* spp. Those which born on little pegs on surface of spore bearing hyphae are called as *redulaspore* (eg. *Botrytis* sp, *Ramularia* sp). *Aleurospore* are the spores born on blown out ends of hyphal tip or by lateral protrusions cut off by septum (eg) *Trichothecium* sp. Single or whorl of thickwalled spores formed through one or more small pores in sporophore wall are called as *porospore* (eg) *Alternaria* sp, *Curvularia* sp. and *Helminosporium* sp. Very often the arthrospheres formed due to breaking up of hyphae are also called as conidia/oidia (eg) *Oidium* sp.

Conidia are of various shape, size, uninucleate to multinucleate, unicellular to multicellular, without pigments or with pigments ranging from light olive to deep black. The conidiophore may be free, simple branched and distinct from each other or may be aggregated to form compound sporophore called as fruiting bodies.

The asexual fruiting bodies produced by the fungi are.

1. Synnemata
2. Sporodochia
3. Acervuli
4. Pycnidia
5. Sorus

## **1. Synnema (Pl . Synnemata)**

It is nothing but the loose aggregation of erect conidiophore so as to form a dense fascicle, similar to mycelial strand. It may split in different ways near the apex, sometimes resembling a feather duster. Such arrangement is called as *coremium* and the conidia are produced at its apex (eg) *Ceratocystis* sp, *Graphium* sp.

## **2. Sporodochium (Pl. Sporodochia)**

It is a fruiting body peculiar to the fungi *Fusarium* sp. It is a cushion shaped aggregation of hyphae which breaks through the host surface and bears conidiophore. These structures may also be formed in the mass of hyphae lying superficially over the substrate.

## **Acervulus (Pl. Acervuli)**

Most of the fungi belonging to the order melonconiales of the deuteromycotina sub division produces acervuli. It is nothing but a saucer shaped depressed mass of aggregated hyphae bearing conidiophore in a compact layer on its exposed surface. In between the conidiophore, long pointed dark coloured structures called *setae* are present. On the host, acervuli are initially developed below the cuticle or epidermis and become erumpent on maturity. (eg) *Colletotrichum* sp, *Pestalotia* sp.

## **Pycnidium (Pl. Pycnidia)**

It is a hollow, flask shaped or globose fruiting body with a narrow mouth (*Ostiole*) whose pseudo parenchymatous inner walls (*Peridium*) are lined with conidiophore which bear conidia (eg) *Macrophomia phaseolina*, *Botrydiplodia theobromae*, *Diplodia natalensis*.

## **Sorus (Pl. Sori )**

Sorus in Greek means heap. ie., the spore bearing hyphae are grouped into small to large masses or clusters (eg) smut sori, rust sori.

## II. Sexual reproduction

This process involves the union of male and female nuclei or nuclei of opposite sex potential. The sex organs are called as gametangia and sex cells are known as gametes. If the gametangia as well as gametes are indistinguishable, into male and female then they are respectively called as *isogametangia* and *isogametes* if distinguishable then called as *heterogametangia* and *heterogametes* respectively. Former one (isogametes) are noticed in most of the zygomycotina fungi while latter type occurs in mastigomycotina and ascomycotina fungi.

The sexual spores are produced mostly at the end of life cycle of the fungi and sometimes in the middle also. Sexual spores function as resting spore in many fungi but in some fungi they do not have any resting period. Accordingly they may or may not have dormant period before germination. The process of sexual reproduction involves three phases.

**1. Plasmogamy** : Union of two protoplast bringing the haploid nuclei of two opposite sex potential close together within the same cell, ie dikaryon.

**2. Karyogamy** : Second phase in sexual reproduction. It is nothing but fusion of two nuclei which are compatible or of opposite sexes. The fused cells are called as zygote. It may occur immediately after plasmogamy such as in lower fungi or it may be considerably delayed such as in the higher fungi. It results in the formation of diploid nucleus.

### Meiosis

It is the third phase in which the fertilized egg (zygote) undergoes the meiotic division. Meiosis reduces the number of chromosome to haploid (n). The nuclei get surrounded by a small amount of the cytoplasm and secrete a wall to become a spore.

Sexual spores are variously known as oospore, zygosporre, ascospore and basidiospore which are produced respectively by mastigomycetina, zygomycotina, ascomycotina and basidiomycotina.

### **Types of sexual anastomosis during plasmogamy (or) fertilization (or) methods of sexual reproduction**

The various methods by means of which compatible nuclei are brought together in the process of plasmogamy are discussed here under.

#### **1. Planogametic coapulation**

Motile gametes are in general called as planogametes. Planogametic coapulation involves the fusion of two naked gametes of which atleast one must be motile gamete. This method is very common in aquatic fungi. If the gametes are morphologically indistinguishable then process of reproduction is called isogamous fertilization (eg) *Synchytrium* sp. If they are morphologically similar but differ in size are called as anisogametes and the fertilization is called anisogamous fertilization (eg) *Blastocladiales*. Among the two gametes, one is small and motile and the other is large and non motile then they are called as heterogametes. The process involving such gametes are called as heterogamous reproduction (eg) Monoblephariales and many higher fungi.

#### **2. Gametangial contact**

This involves fusion of two gametangia which are morphologically distinguishable with undifferentiate protoplasm, each having one nucleus for fusion though many come in contact with each other. One important difference from the previous method is that the gametes are never released from one gametangium into the other.

In this method, two gametangia of opposite sex come in contact and one or more gametic nuclei move from the male gametangium to the female, through a pore developed by the dissolution of gametangial wall at the point of contact (eg) *Pythium* sp. while in some fungi, a specially developed fertilization tube (*trichogyne*) act as a passage for the male nuclei to enter into female gametangium. After the passage of male nuclei, male gametangium starts to disintegrate and female gametangium grow continuously to form spore. (eg) Ascomycotina fungi.

### **3) Gametangial copulation (or) aplanogametic copulation**

The difference from the gametangial contact lies in only passing the entire contents of one gametangium to another gametangium instead of passage of only the nuclei. Here fusion may takes place in two ways.

1. The entire contents of one gametangium passes into another through a pore developed on the wall at the point of contact (eg) *Chytridiomycetes* (holocarpic fungi).
2. Two morphologically similar gametangial hyphae come in contact, the wall at the point of contact dissolve, resulting in a common cell in which the protoplast mix. This type of plasmogamy is otherwise called as *isogamous conjugation* (eg) *Mucor* sp.

*Rhizopus* sp.

### **4) Spermatization**

This method is commonly found in only ascomycotina and basidiomycotina. Some fungi produces numerous, nonmotile minute, uninucleate spore like structures called as *spermatia* in a special sexual apparatus called as *spermagonium*. This spermatia behave as male gamete and are usually carried by insects, wind and water to the female sex organ viz., receptive hyphae or even to the somatic hyphae itself. On dissolution of cell wall at the point of contact the entire content of spermatium pass into female sex organ.

## **5. Somatogamy**

No sex organs are produced but the somatic cells function as gametes, a common method of reproduction in higher ascomycotina and basidiomycetes and is virtually absent in lower fungi. In this process, somatic hyphae anastomose through clamp connection and the nuclei of opposite mating type are brought together in one cell. This dikaryotic cells eventually undergoes karyogamy and meiosis to develop sexual spores.

The sexual spore of fungi is of two kinds.

1. Diploid resting spore (eg) Oospore, Zygospore
2. Haploid non resting spore (eg) ascospore, basidiospore.

### **1 Oospore**

They are the resting diploid sexual spores produced by the members of Oomycetes, through gametangial contact. The male gametangium is known as anthredium and female one is called as Oogonium.

Anthredium is an elongated, club shaped, multinucleate cell, much smaller in size than oogonium and are produced terminally (or) intercalary. There are different types of anthredium.

#### **a) Monoclinous**

Anthredium may arise anywhere along the Oogonial stalk (eg) *Pythium* sp.

#### **b) Diclinous**

Anthredium arise from another branch of thallus (eg) *Pythium* sp.

#### **c) Paragynous**

Tip of the anthredium is attached parallel sto the side of oogonium during gametangial contact (eg) *Pythium* sp, *Albugo* sp.

#### d) Amphigynous

Oogonium perforate the walls of anthredium and penetrate into it finally emerging out at the other side as a spherical structure. While passing through anthredium, the fertilization is effected. On maturity, the sexual apparatus consist of a balloon shaped Oogonium with a thick walled Oospore at the top and a funnel shape base formed by the anthredium. (eg) *Phytophthora* sp.

The female sex organ, Oogonium is a globose structure which develops from a terminal or intercalary swellings of the hyphae. The uninucleate dense central region of oogonium is called as ososphere ooplasm and an outer or peripheral multinucleate spongy portion lining oogonial wall is called as periplasm.

Oospore formed by the fusion of anthredium and oogonium have an outer most thick wall (exine), inner thin delicate wall (intine) with a central oil globules (Food reserve). Exine may be smooth or rough according to the species.

The oospore may be pleurotic / apleurotic. If the oospore completely fill the oogonial cavity then it is called as *pleurotic* (eg) *Pythium* sp. and those which does not fill Oogonial cavity are called as *apleurotic* (eg) *Albugo* sp., *Phytophthora* sp. But in some members of the lower fungi, Oosphere failed to get fertilized and oospore develop parthenogenetically. For example in *Saprolegnia fexae* no anthredia are formed and the eggs of oogonia itself function as parthenospore and behave similar to oospore.

The oospores after a period of rest it may germinate directly indirectly and give rise to mycelium. In case of direct germination oospore germinate into mycelium without any intermediate stage of development, after period of rest (eg) *Pythium* sp, *Phytophthora* sp. Whereas the oospore of *Albugo* sp, after a period of dormancy germinate and release zoospore first which inturn swim for sometime, encyst and germinate to mycelium.

## 2) Zygospore

The resting, diploid, sexual spore of the class zygomycetes are called as zygospore which are obtained through isogamous gametangial copulation of two compatible hyphae.

## 3) Ascospore

Sexual spore of ascomycotina are called as ascospore. These haploid spores are formed by either one of the following methods.

1. Gametangial contact
2. Gametangial copulation
3. Spermatization
4. Somatogamy

Out of these four methods, gametangial contact is noticed in many members of the ascomycotina. The sex organs are anthredium (male) and ascogonium (female). Another important feature in the development of ascospore is that the karyogamy is not effected immediately after plasmogamy and the fused gametes (zygotes) first undergo meiosis resulting in 4 haploid nucleus and then each nuclei divide mitotically to form 8 nuclei. Each nuclei is enveloped by a wall to form ascospores which are present inside the sac like structures called *ascus* (Pl. *Asci*). Asci are generally produced inside the fruiting body called as ascocarp. The ascocarp may be cleistothecium (completely close ascocarp) perithecium (closed with a pore) apothecium (open ascocarp). But in *Taphrina* the asci are not produced inside the ascocarp (naked asci).

## 4. Basidiospore

Sexual spore produced by basidiomycotina are called as basidiospore. Somatogamy and spermatization are the two methods by which plasmogamy occurs.

Basidiospore are produced at the tip of four sterigmata present on a club shaped structure called as basidia.

Basidiospore may or may not be produced inside the fruiting body called as basidiocarp. Usually higher basidiomycetes produce their basidia on fruiting bodies than lower basidiomycetes.

## **LECTURE NO.6,7,8,9&10**

### **GENERAL SYMPTOMS OF FUNGAL DISEASES**

#### **1. Damping off**

- Rotting of seeds and radicle before the seedling emerge out of the soil (pre-emergence damping off)
- Rotting of emerged seedlings at ground level resulting in toppling down of seedlings. (Post – emergence damping off).

Eg: Damping off of Vegetable - *Pythium aphanidermatum* *P. debaryanum* *P. ultimum*

#### **2. Club root**

Modification of roots and rootlets of plant into small or large spindle like spherical, knobby or club shaped swellings are called as club root.

Eg: club root of cabbage : *Plasmodiophora brassicae*

#### **3. Root rot**

Disintegration or decay of roots

Eg: Root rot of pulses *Macrophomina phaseolina*

#### **4. Foot rot**

Disintegration of basal part of the stem

Eg: Foot rot in pepper *Phytophthora palmivora*

#### **4. Collar rot**

The Collar region of the seedlings rot

Eg: Collar rot of groundnut – *Aspergillus niger*

*A.pulverulentus*

**5. Stem rot :** leads to rotting

Disintegration of the tissues of the stem.

Eg : Stem rot of paddy – *Sclerotium oryzae*

**6. Wilt**

Loss of turgidity and dropping of leaves and shoots

Eg: Red gram wilt – *Fusarium oxysporum* f.sp. *udum*

**7. White rust**

White or creamy, shiny irregular pustules seen on the lower surface of the leaves.

Eg: White rust of amaranthus - *Albugo bliti*

**8. Downy mildew**

- \* Mildew growth seen on the lower surface of the leaf
- \* Corresponding upper surface are chlorotic

Eg: Cumbu downy mildew – *Sclerospora graminicola*

**9. Powdery mildew**

- \* Small irregular powdery patches are seen on the upper surface of the leaves.

Eg: Powdery mildew of blackgram – *Erysiphe polygoni*.

**10. Leaf spot**

- \* Localized necrotic lesions on host leaves.
- \* The spots vary in size, shape and colour

**11. Leaf blight**

Rapid browning and death of leaves giving a burnt appearance

Eg: Leaf blight of sorghum – *Exerohilum turasicum*

**12. Blast**

- \* Appearance of spindle shaped spots with grey centre and brown margin on leaf (leaf blast)
- \* Blackening of node of the plant (node blast) and neck of the panicle (neck blast)

Eg: Blast of paddy – *Pyricularia grisea*

### **13. Anthracnose**

- \* Black sunken lesion circular to angular are seen on leaves, cotyledons, stem, fruits, flowers and pods of plants.
- \* Acervuli the asexual fruiting body is produced in the sunken lesion.

Eg: Anthracrose of bean – *Colletotrichum lindemuthianum*.

### **14. Rust**

- \* Raised reddish brown (rusty) pustul appear on both surface of leaf, stem etc

Eg: Sunflower rust - *Puccinia helianthe* :

### **15. Scab**

Slightly raised or sunken lesion with cracks on petiole, pedicel, blossom, fruit, leaves and gives a scabby appearance.

Eg. Scab of apple - *Venturia inaequalis*

### **16. Sooty Mould**

Superficial dark black sooty growth found on leaf, stem and fruits.

Eg: Sooty mould in mango – *Capnodium mangiferae*

### **Gummosis**

- \* Exudation of gummy substances from infected bank.
- \* The bark completely rots, and tree dries due to girdling

Eg: glummosis of citrus – *Phytophthora citrophthora*

### **18. Sugary disease or Ergot**

- \* Exudation of creamy, sticky honey dew like secretion from the infected floret (Sugar disease).
- \* Long, straight or curved cream to dark brown hard sclerotia replaces the grain in the ear head (Ergot)

(Eg) Cumbu – *Claviceps fusiformis*

### **19. Smut**

Seeds in the infected ear head or entire earhead is replaced by a gall (or) sori contain large number of black colour , smut spores or spore bolls.

(eg) Loose smut of wheat – *Ustilago nuda tortice*

## **20. Fruit rot**

- \* Rotting of fruit.
- \* The infected portion is converted with mouldy growth.  
(Eg) Fruit of citrus : *Penicillium expansum*

## **21. Die back**

Drying of twigs from the tip downward (eg) die-back of mango *Botryodiplodia theobromae*.

## **22. Bunt**

- \* The infected grain are found to be full of sooty black powdery masses of bunt spores decayed fishy odour and emits a smell) Bunt of wheat *Tilletia caries*.

## **23. Wart**

Wart like protuberances on tubers and stem (eg) wart of potato *Synchytrium endobioticum*.

## **24. Soft rot**

Maceration and disintegration of root, bulbs, tubers, fruits and fleshy leaves.

## **25. Gall**

Enlarged sac like growth on stem, leaves, blossoms etc.

(eg) Stem gall in coriander (*Protomyces macrosporus*).

## **26. Leaf curl**

Distortion, thickening and curling of leaf.

(eg) Leaf curl of peach – *Taphrina deformans*.

## **27. Chlorosis**

Loss of chlorophyll giving pale green appearance to the green plant parts.

## **28. Necrosis**

Killing of plant tissue often results in development of brown to black colour.

## **29. Discolouration**

Change in colour of the plant or one or more its parts.

## LECTURE NO.11

### GENERAL CHARACTERS OF MASTIGOMYCOTINA

- Produce flagellated cells during their life cycle.
- Majority of them are filamentous coenocytic mycelium .Some times pseudosepta produced. Some bear rhizoid.
- Centric nuclear division occurs. Centrioles remain functional during nuclear division.
- Absorbitive type of nutrition.
- Produce zoospores.
- Oospore are the sexual spores

### GENERAL CHARACTERS OF ZYGOMYCOTINA

- Majority of them are saprobic some are coprophilous some are weak parasites attacking plants.
- It produces well developed branched coenocytic mycelium.
- Cell contains mitochondria ,nucleus,ribosomes,lipid granules and endoplasmic reticulum Cell wall composed of chitin
- Centrio les are absent
- Asexual reproduction by sporangiophores
- Reproduce by producing cWamydospores ,few produce oidia
- Zoospores are absent
- Sexual reproduction is by gametangial fusion
- Gametangial fusion results in thick walled resting zygospores

## LECTURE NO. 12

### GENERAL CHARACTERS OF ASCOMYCOTINA

- Mycelium is well developed ,branched ,septate. Yeast is single celled organism
- Septum has a central pore
- Cell wall is made up of chitin .Chitin is in the form of microfibbrillar skeleton in filamentous organism.
- Ascus is usually sac like and contains usually 8 ascospores.
- Ascospores are endogenous in origin
- Ascocarps are of different shapes
- Apothecium in discomycetes
- Flask shaped perithecium in Pyrenomycetes

- Closed cleistothecium in Plectomycetes
- No flagellated cells
- Asexual spores are conidia

## **Ascomycotina**

### **General characters**

1. The ascomycetous fungi exhibits wide range of habita, it may be coprophilous growing on dung or marine or fresh water. Powdery mildews Ectoparasites. Few are hypogean (develop under ground).
2. The mycelium is well developed, profusely branched and septate. Yeasts are single celled organisms.
3. In each septum or crosswall of the mycelium there is present a simple central pore which allows the mitochondria, nucleic and other cytoplasmic contents to pass from cell to cell.
4. The cell wall contains chitin
5. The sexual fruiting body is the ascocarp which remain enveloped in a sheath of sterile hyphae. The ascocarps are of different types.

Saucer shaped Apothecium (e.g) Discomycetes

Flask shaped perithecium - Pyrenomycetes closed, spherical and indehiscent -- cleistothecium -- Plectomycetes

A sexual reproduction takes place by the non-motile spores called conidia, pycniospores, oidia or chlamydospores.

### **Ascus Development**

Ascus develops after fertilization by direct or indirect method.

### **Direct development of ascus**

In lower Ascomycetes plasmogamy is followed immediately by Karyogamy resulting in the formation of a diploid nucleus. The cell containing this diploid nucleus develops directly into an ascus. This nucleus divides first meiotically and then by ordinary divisions, resulting into eight haploid nuclei, which ultimately change into eight ascospores (eg) *Saccharomyces*, *Dipodascus* etc.

### **Indirect development of ascus**

The two gametangia (antheridium and ascogonium) come in contact with each other. The male nuclei form the antheridium pass through the trichogyne into the ascogonium and get themselves paired with the female nucleic and this paired nucleic are called dikaryons. At this stage many dikaryons are present in the ascogonium. The wall of ascogonium gives rise to many papilla like outgrowth as which devlop into short ascogenous hyphae. The apired nucleic or dikaryons migrate into these ascogenous hyphae young ascogenous hyphae are aseptate, but later on they become septate. Of these two nuclei of the cells of ascogenous hyphae one is of antheridial origin and the other is of ascogonium develop many sterile hyphae to form pseudo parenchymatous hollow apparatus. Some of the sterile hyphae form a thick peridium around the ascocarp and many develop into paraphyses in the fruiting body.

The asci develop at the tips of ascogenous hyphae. The terminal cell of each ascogenous hypha, contains two nuclei. It curves and forms a crozier or hook. Both the nuclei of crozier divide simultaneously to form four nuclei. Septum forms in such a way that a uninucleate tip cell, binucleate penultimate cell and a uninucleate basal cell are formed. This penultimate binucleate cell becomes the ascus mother cell. Of the two nuclei in this ascus mother cell one is of antheridial origin and other of ascogonial origin. It enlarges and develops into an ascus.

The two nuclei of this ascus mother cell fuse and form a diploid nucleus. This diploid nucleus divides first meiotically and then ordinarily or mitotically, forming four haploid nuclei. All these nuclei get surrounded by some cytoplasmic contents and change into eight ascopores.

## **LECTURE NO. 15,16,17,18,19,20,21,22,23 & 24**

### **Systematic position**

Division - Myxomycota  
 Class - Plasmociliophoromycetes  
 Order - Plasmodiophorales  
 Family - Plasmodiophoraceae

### **Symptoms**

1. The dropping of leaves at noon or bright days is the first sign of symptom.  
Subsequently it leads to permanent wilting or retarded growth
2. Roots are hypertrophied and enlarged to form club shaped malformation and hence the disease is known as club root of cabbage.
3. The late infection produce galls on the lateral branches suggesting an alternative name and 'Finger and Toe' disease.

### **Life cycle**

#### **Primary phase**

The resting spores are tiny, hyaline spherical, uninucleate with spiny walls. Infected host roots get decayed and the resting spores are released into the soil. The spores germinate immediately with the help of the root exudates of the host or remain dormant for several years. During germination a pore is dissolved on the cyst wall through which an apically biflagellated spherical or pear shaped uninucleate, naked mass zoospore comes out.

1. The zoospore gets attached to the host
2. Inactivation of its flagella, retraction of axonemes and encystment of the zoospore takes place.

3. A tubular cavity, rohr appears within the excepted zoospore and the end is plugged with light staining plug. The rohr forms a bulbores structure and is attached to the host with the hupot adhesorium
  4. A dark staining rod is present within the tubular robr calla stachd. This punches the host cell wall and the contents of the zoospore enters the host cell and is spherical and amoeboid.
- This amoeboid zoospores are called myxamoebae.
  - Plasmogamy of these haploid amoeboid protoplast result in the formation of multinucleate primary plasmodia.
  - Later by clevage, multinucleate segments are delimited each of which develops into a zoosporangium. Thus the plasmodium becomes a sonus consisting of several sporangia.
  - Each sporangium form 4-8 uninculeate anteriorly biflagellate zoospores. Exit tubes originate from the sporangia or a pore develops between the zoosporangium and the cell wall and through this pore the zoospores are liberated. The zoospores function as gametes and these (isogamy) and form a binucleate of vadriflagellate motile zygote.

### **The secondary phase**

The zygote settles on the root have, sheds the flagella and enters the cortex of the root cell.

- ❖ Each of the binucleate plasmodium enlarges and undergo repeated mitotic nuclear division to form a multinucleate body which is referred as multinucleate secondary plasmodium. So far karyogamy does not take place.
- ❖ The root cell containing the multinucleate plasmodia get hypertrophid and takes the shape of a club.

- ❖ The haploid nuclei get associated in pairs Kanjogamy takes place and diploid zygotic nucleic are formed.
- ❖ Meiosis takes place soon after the formation of diploid aggote. The diploid mulchi of the plasmodium at the time of spore formation pass through a special phase called akanjote phase or stage. During the phase the nuclei seem to disapper during the prophase of meiosis the nucleic reappear.
- ❖ It is followed by nulotic division resulting in haploid nucleic by cleavage of the cytoplasm and wall formation several haploid resting spores are formed. on being liberated these resting spores are able to reinfert the host.

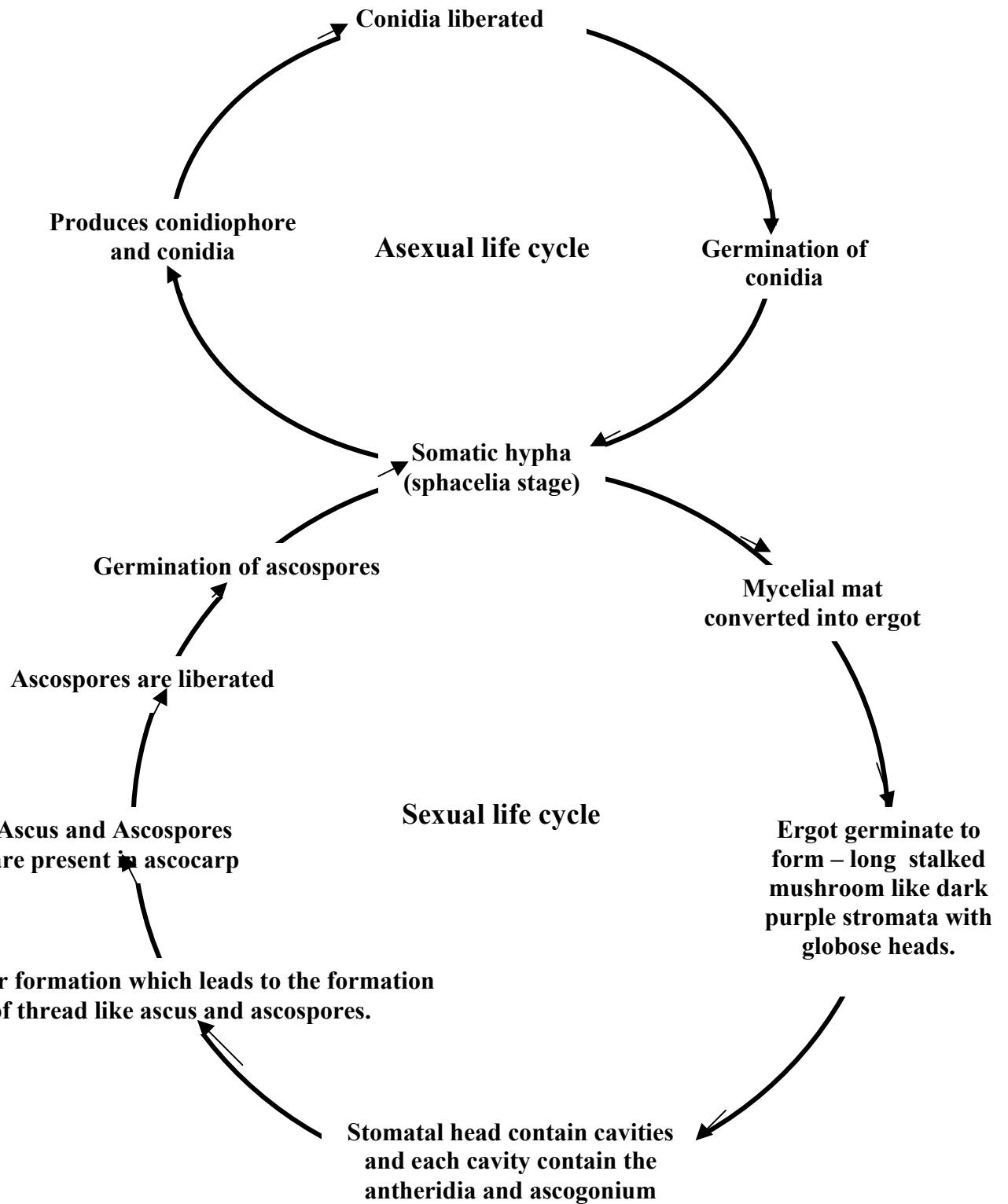
### **Alternation of generations**

There is a distinct alternation of generation in the lifecycle of *Plasmodiophora*. The zygote and the diploid plasmodium represent the sporophyte phase. The resting spores, the zoospores, the haploid plasmodium and the isogamety represent the gametophyte phase.

With meiosis the diploid or sporophyte phase ends and the gametophyte phase starts. At the time of Kanjogamy the sporophyte phase ends and the sporophyte phase is initiated with the formation of diploid zygote.

### Life cycle of *Claviceps purpurea*

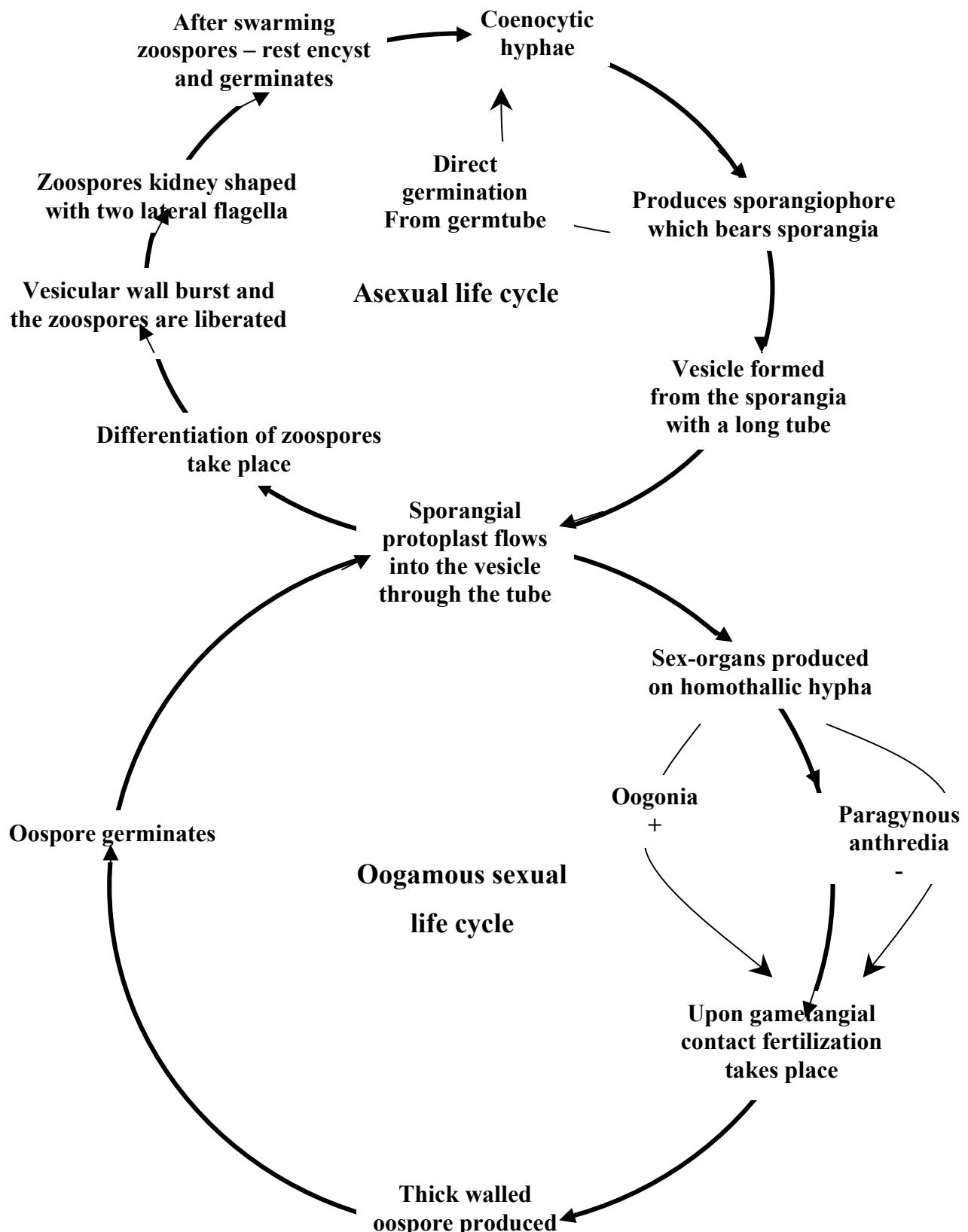
Disease-Ergot or sugary disease of cumbu.



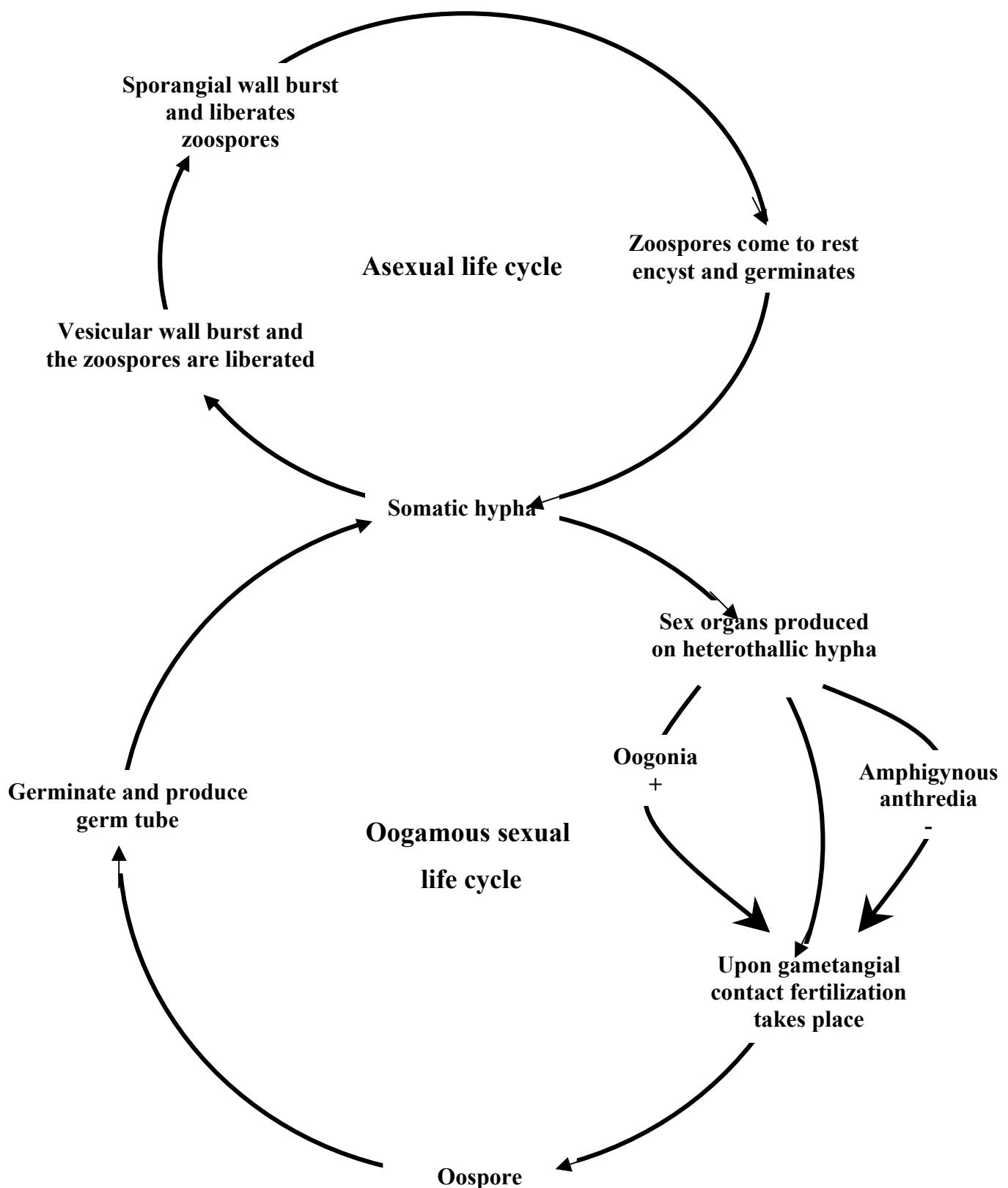
**Life cycle of *P. debaryanum***

<b>Kingdom</b>	:	<b>Protista</b>
<b>Sub – Kingdom</b>	:	<b>Mycota</b>
<b>Division</b>	:	<b>Eumycota</b>
<b>Sub-Division</b>	:	<b>Mastigomycotina</b>
<b>Class</b>	:	<b>Oomycetes</b>
<b>Order</b>	:	<b>Peronosporales</b>
<b>Family</b>	:	<b>Pythiaceae</b>
<b>Genus</b>	:	<b>Pythium</b>
<b>Species</b>	:	<b>debaryanum</b>

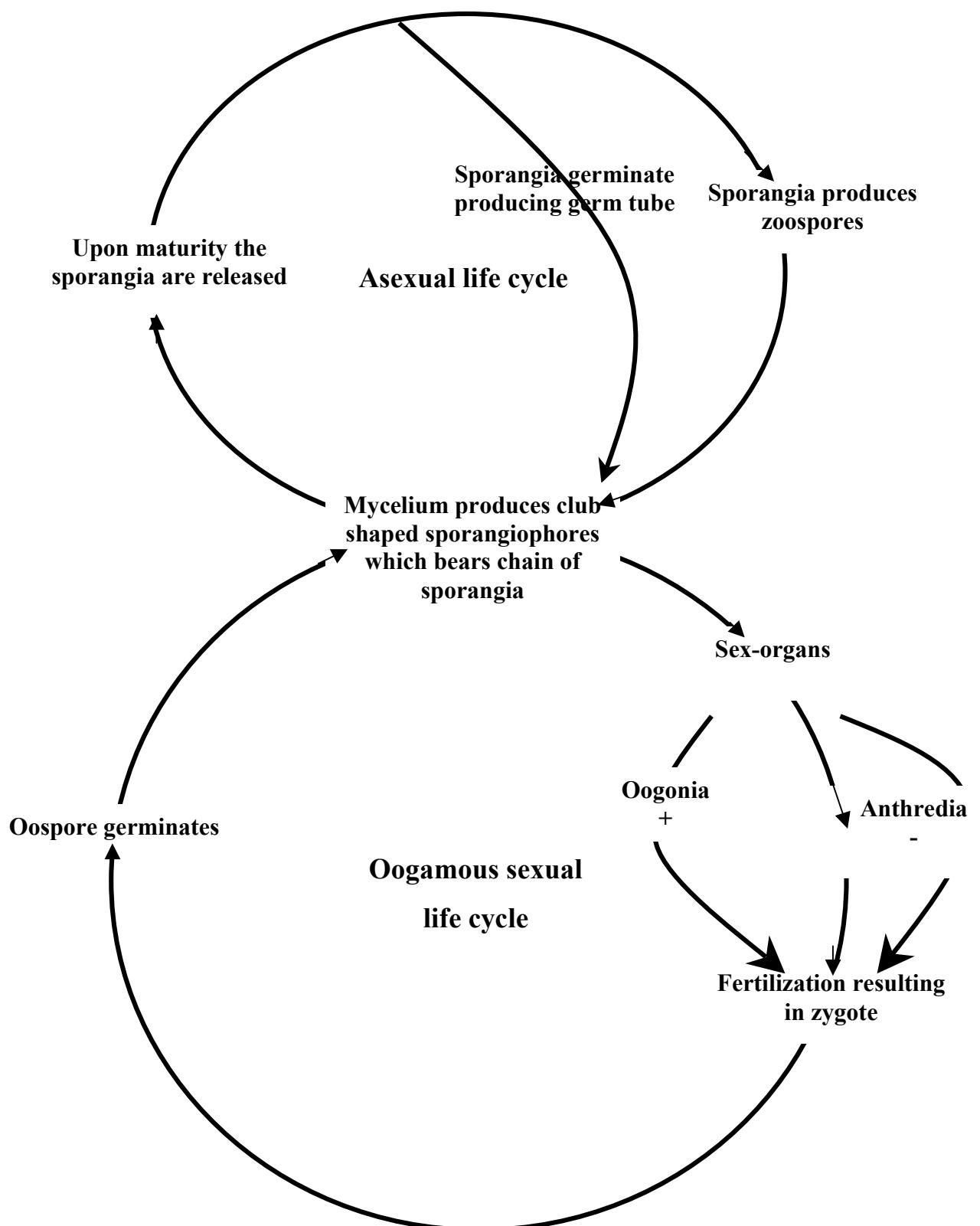
### Life cycle of *P. debaryanum*



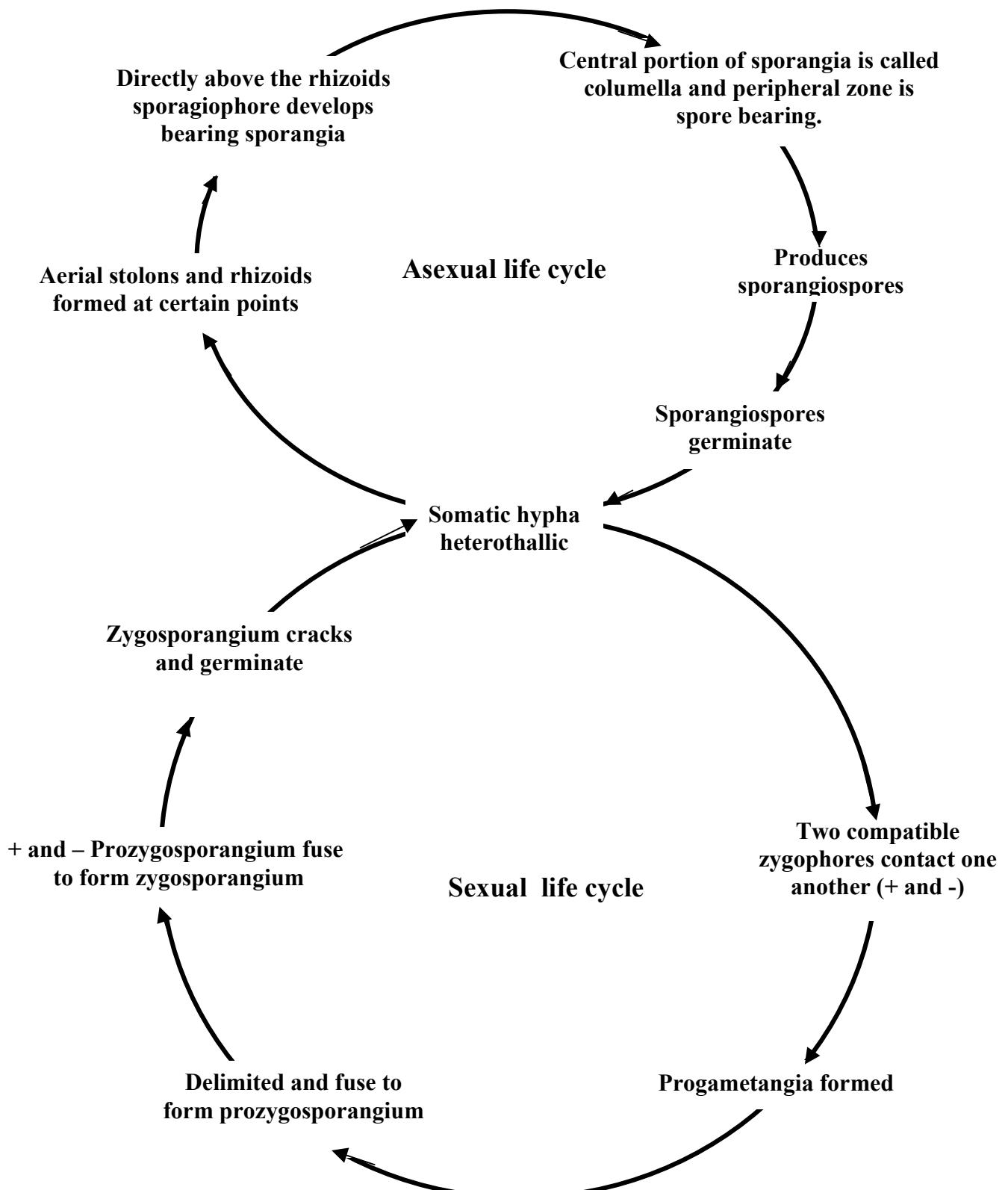
### Life cycle of *Phytophthora infestans*



### Life cycle of *Albugo candida*



### Life cycle of *Rhizopus stolonifer*



## LECTURE NO.19

### **Life cycle of *Rhizopus stolonifer***

#### **A sexual reproduction**

The sporangiophores are erect and produce pin head like black coloured sporangia. Unicellular, multinucleate, non motile spores are produced in the sporangia. Due to the pin head like black coloured sporangia the entire mycelium appears blackish and hence the popular name 'black mold'.

The sporangiophore swells at the tip into a knob like vesicle and the cytoplasm along with many nuclei flows into the swollen vesicle, which represents the young sporangium. The protoplasm in the sporangium become differentiated into two zones, i.e. outer peripheral multinucleate dense region and the central less denser region with comparatively fewer nuclei. The two portions get separated by a layer of vacuole and a wall is secreted in this region, thus a outer sporangiferous zone and central columella is formed in the sporangium. Clevage in the peripheral sporangiferous zone results in the formation of many multinucleate segments which secrete wall around each of them and form unicellular, globose or oval multinucleate, non-motile sporangiospores called aplonospores. The sporangial wall and the columella breaks and the dry spores are liberated in the air. The spore germinates by producing a germ tube, that develops into a fluffy, well branched white aerial mycelium. *R. stolonifer* rarely produce chlamydospore.

#### **Sexual reproduction**

*Rhizopus* reproduces sexually by the process of conjugation which results in the formation of zygospores. *Rhizopus stolonifer* is heterothallic and *R. sexualis* is homothallic. In this heterothallic species the two fusing mycelia belong to two different mating types, + strain and - strain.

### **Telemorphotic reaction**

At the time of sexual reproduction initiation of club shaped, aerial zygomycete is formed. The zygomycetes in both the strains + and - are induced by trisporic acids B and C. Under zygotropic reaction the zygomycetes of opposite strains approach towards each other and from each develops a copulation branch. The two copulating branches are called progametangia. The progametangia adhere together at their tips and the cytoplasm and nuclei flow into the progametangia and the zygomycetes are pushed apart. The tip of each progametangium is soon cut by a septum. The small terminal cell so formed is called gametangium whereas the long tubular part is called a suspensor. The gametangium has dense granular multinucleate protoplast whereas the suspensor has a more vacuolated protoplast. A large pore develops in the adjoining wall of the two gametangia which allows both the gametangial protoplasts to fuse and form a zygosporangium. The zygosporangium soon becomes surrounded by a thick, black, warty wall. The zygosporangium wall is made up of two layers, of which the outer dark thick and warty layer is called exine and the inner thin layer is called intine. After a period of rest, meiosis takes place at the time of zygosporangium germination. On germination a lateral crack is formed on its wall. The inner thin intine comes out in the form of a hypha like germ tube, which is also called promycelium. The meiotic divisions results in a number of haploid nuclei in the protoplasm of germinating zygosporangium. The young germ tube functions as a sporangiophore and develops a germ sporangium at its tip. Many germspores or meiospores remain filled in this sporangium. The germ sporangia of *R. stolonifer* contain either entire + or entire - spores or a mixture of both. These spores germinate to form fresh mycelium.

### **Differences of *Rhizopus* from *Mucor*.**

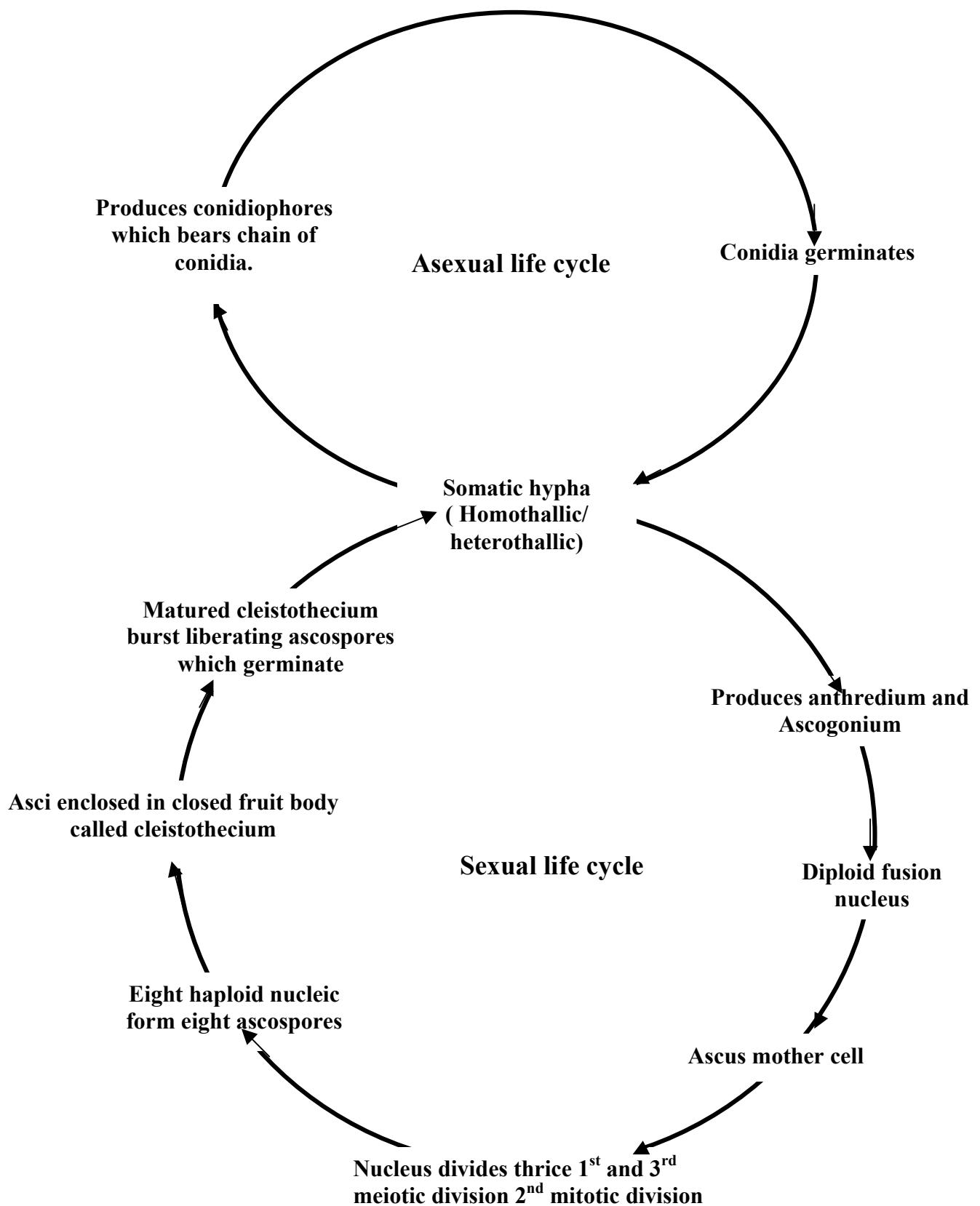
<b>Sl.No.</b>	<b>Rhizopus</b>	<b>Mucor</b>
1.	Rhizoids (absorptive hyphae) or holdfasts are present	Rhizoids or holdfasts are generally absent, or less specialized
2.	Stolons are present	Stolons are absent

3.	Food material is absorbed by rhizoids	Food is mainly absorbed by the entire mycelial surface
4.	Sporangiophores develop in well organized groups mainly against the rhizoidal hyphae	Sporangiophores arise singly, and not in groups
5.	Spores remain adhered to columella and are not easily disseminated	Spores easily blown away by wind.

## LECTURE NO.20

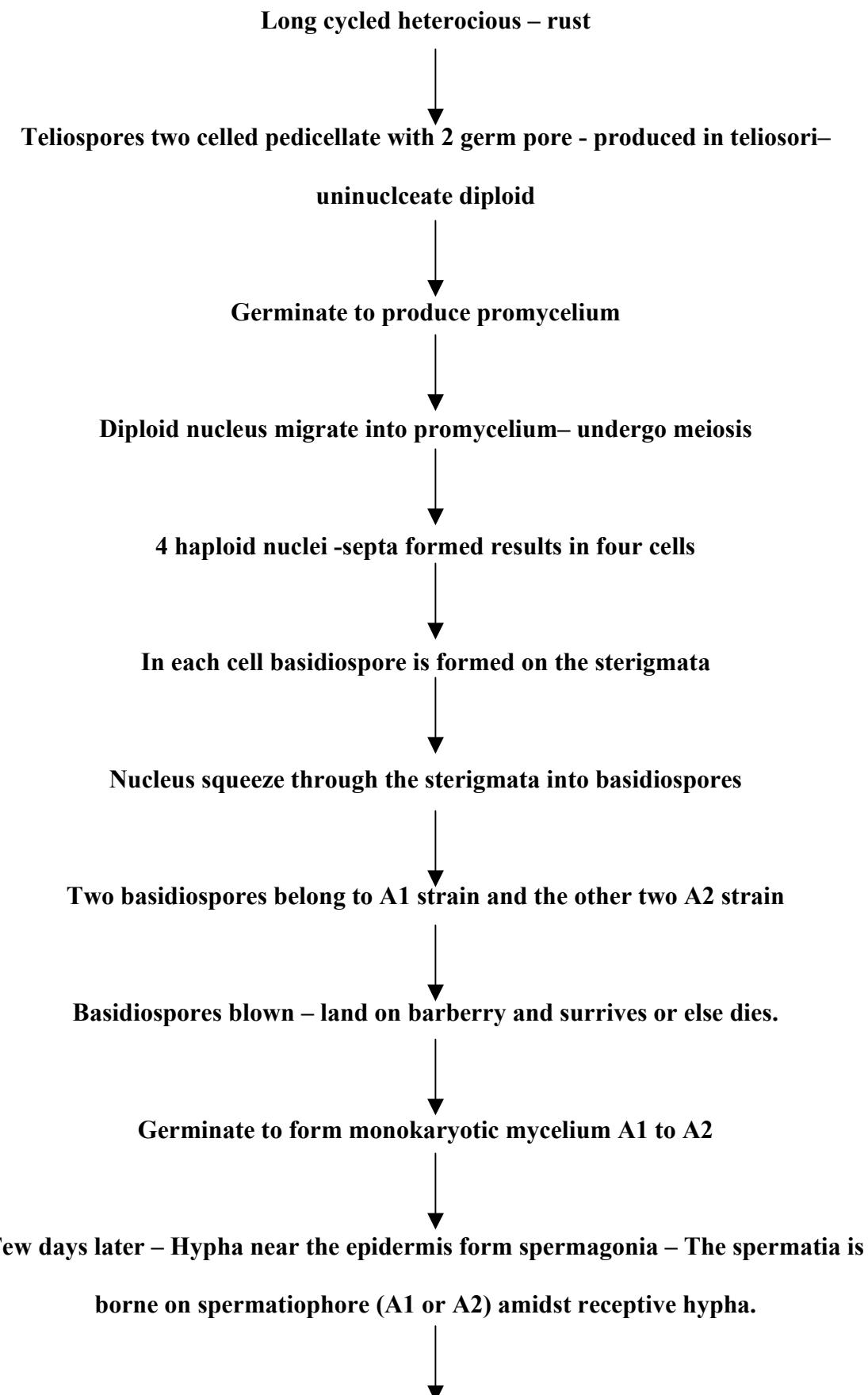
Life cycle of *Erysiphe*

Disease -powdery mildew of greengram, blackgram, bhendi



## LECTURE NO.21

### Life cycle of *Puccinia*



**Spermatization takes place –A1 spermatia is transferred to A2 by insects**



**Mycelium permeates the entire leaf reaches the lower epidermis and forms aecial primordia.**



**During spermatization nucleus pass from spermatia into receptive hypha travel down the hypha through septal perforation and reach aecial primordia rendering it binucleate – First binucleate spore.**



**Dikaryotization is followed by formation of aecial cup with aeciospores (spherical, smooth, rough walled and binucleate)**



**Aecial cup breaks open through the lower epidermis and the aeciospores fall on wheat**



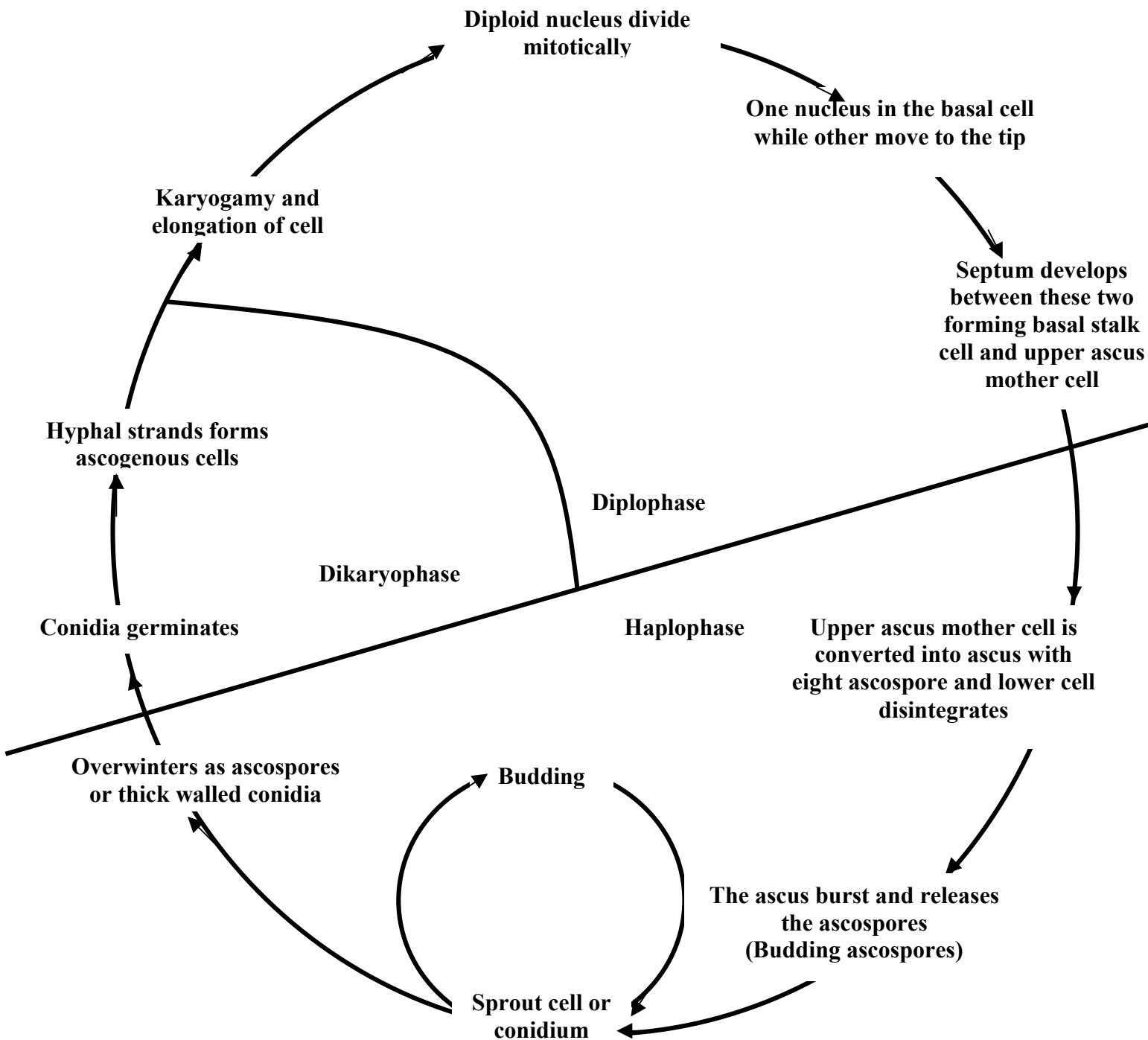
**The binucleate mycelium forms the uredinia with urediospore. They are**

**repeating spores which help in the perpetuation of the fungus.**

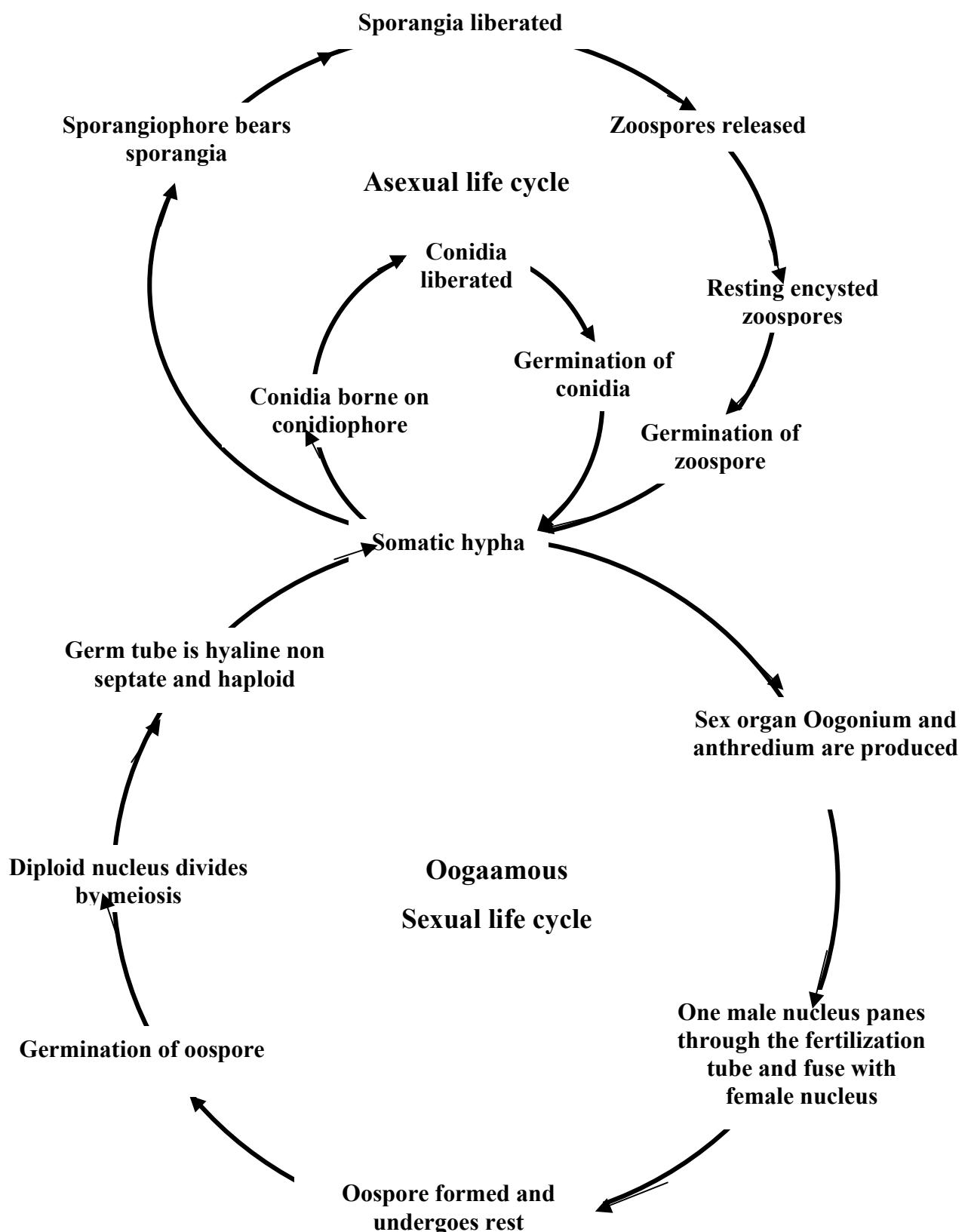


**Teliospores are formed and the cycle is repeated.**

### Life cycle of *Taphrina deformans*



### Life cycle of *Sclerospora*



## LECTURE NO.25

### Plant bacteriology

#### Introduction

Bacteria are unicellular, microscopic, chlorophyll less, prokaryotic microorganism reproduce mainly by fission. Existence of bacteria in the ecosystem was discovered first by the Dutch man, *Anton von Leeuwenhoek* in the year 1675 from Holland with the help of high power lenses. In 1876, Pasteur and Koch of France for the first time proved that anthrax disease in animal was caused by a bacterium. The association of bacteria with plant disease was first showed by T.J. Burill of USA in 1882. He actually identified that fire blight disease of pear was caused by a bacterium and now it is known as *Erwinia amylovora*.

About 180 species of bacteria are known to cause diseases in crop plants. Most of the plant pathogenic bacteria are facultative saprophytes and can be grown artificially on nutrient media, however some fastidious vascular bacteria are difficult to grow in culture. The most important botanical families attacked by bacteria are the gramineae, solanaceae, leguminaceae and Rosaceae.

#### General account of bacteria

Bacteria are primitive prokaryotic microorganism with a rigid cell wall and their nuclear material is not separated from the cytoplasm by a nuclear membrane and there is no mitotic apparatus.

#### Types of bacteria

Majority of the bacteria measures approximately  $0.5 - 1.00 \mu\text{m} \times 2.0 - 5.0 \mu\text{m}$ . The shape of the bacterium is governed by the rigid cell wall. Usually bacteria have three principal shapes (1) Coccus (Pl. Coccii) (2) Bacillus (Pl. Bacilli) and (3) spirillum (Pl. Spirilli).

## **Coccus**

These are spherical in shape, may be arranged singly (micrococcus), in pairs (diplococcus), chains (strapto coccus) in box like cubical poekets (sarcinae), inetrads (Pair of four cells in same plane) or irregular grape like clusters (Staphylococcus).

## **Bacillus**

These are straight rod or cylindrical or ellipsoidal in shape, occur mostly singly, sometimes (microbacilli) in pairs (diplobacilli), in chains (Streptobacilli) or in parallel palisade like arrangements.

## **Spirillum**

These are cork screw shaped i.e the rods that are helically curved with a twist. Rod shape bacteria with less than one complete turn or twist have vibrioid shape where as those with one or more complete turns have a helical shape.

Bacterial cells in the chain have minimum essentially tangential contact between the cells. When the chain consist of cells with substantial contact with adjacent cells than that chain is called as trichome. Some bacteria are pleomorphic and their shape changes to different types in response to change in environment condition.

Most of the plant pathogenic bacteria are rod shaped except spp. of *Streptomyces* which are mostly filamentous which resemble to the morphology of fungi. Many plant pathogenic bacteria have one or more flagella for some limited movement. The position and number of flagella varies with bacterial genera. Accordingly the rod shape bacteria are grouped as

- |                     |   |
|---------------------|---|
| 1. Atrichous        | - With out flagella (eg) <i>Xylella</i> sp.                                 |
| 2. Monotrichous     | - Single flagellum at one end (eg) <i>Xanthomonas</i> sp.                   |
| 3. Cephalo trichous | - Several numerous flagella at one pole (eg) <i>Pseudomonas fluorescens</i> |

4. Amphitrichous - Atleast one flagellum at each pole (eg) *Pseudomonas* spp
5. Peritrichous - Flagella protruding from all portion of its surface (eg) *Erwinia* spp.
6. Lophotrichous - Two or more flagella at both the poles of the bacterium. Most of the spirillum type of bacteria are the common examples.

Gram staining reaction differentiates bacteria into gram positive and gram negative. gram-positive bacteria retain violet iodine stain combination because it forms a complex with certain components of their cell walls and cytoplasm. Gram negative bacteria does not retain stain. Among the plant pathogenic bacteria, *Clavibacter* sp (*Corynebacterium*) and *Curtobacterium* sp. are gram positive whereas *Agrobacterium* sp, *Erwinia* sp, *Pseudomonas* sp, *Xanthomonas* sp and *Xylella* sp. are gram negative.

### **Structure of bacterial cell**

Bacterial cells exhibit a typical prokaryotic structure. The various structures of bacterial cell are discussed in detail hereunder.

#### **1. Flagella (Singular : Flagellum)**

Swimming is one mode of locomotion of bacteria in a liquid environment which is aided by flagella. Flagella are hair like helical structures that protrude through the cell wall and are usually exceeding the length of bacteria. Bacterial flagella are usually very thinner than that of eukaryotes. Individual flagellum composed of a basal body associated with cytoplasmic membrane (*Kinetoplast*) and cell wall; a short hook and a helical filament. Filament is made up of protein namely flagellin. Gram negative bacteria have a sheath surrounding the flagellum.

Motile bacterial cells are capable of directing the swimming position towards (positive) or away negative from various chemical compounds and such phenomena is called as chemotaxis.

In addition to flagellar motility non-flagellated bacteria have gliding motility in solid surface and are mainly due to the involvement of microfibrils (excreted slime) and other similar protrusion of the cell surface. But some sps of *Pseudomonas* sp. exhibit twitching motility on solid media. This is because of presence of thin, polar *pili* or *fimbriae* on the cell surface.

## **2. Pili/fimbriae**

These are short, fine, filamentousl non helical appendages, usually more numerous than flagella extending outward from the cell surface Chemically they are made up of a protein namely *pilin*. They manly play a important role in adhering bacterial cells to the host surface and conjugation. Mostly gram –ve bacteria have pili.

## **3. Capsule**

This is the outermost layer of bacterial cell. Capsule is slimy or gummy or viscous in nature forming a envelop around the cell wall and synthesised by cell membrane.

Capsule offer protection to the bacterial cell against desiccation. If this envelope or covering is too thin then they are called as microcapsule. On contrary, the mucous membrane is so abundant and embedding so many cells as a matrix then they are called as slime.

Capsules are basically polysaccharides with some protein made up of polymers of various subunits and its composition varying with the species. In *Bacillus* sp, it is made up of single amino acid namely glutamic acid and in *Agrobacterium*, the slime layer is purely glucan.

#### 4. Cell wall

The cell wall not only gives shape to the bacterium but also has a vital mechanical function. It is relatively thin, rigid and that allows inward passage of nutrients and outward passage of waste materials and digestive enzymes.

All materials present inside the cell wall is called protoplast. Protoplast is enclosed by a plasma membrane or cytoplasmic membrane.

This membrane actually regulates the passage of materials into and out of the cell and this is the site of various metabolic activities. Exterior to this, there is a layer made up of peptidoglycan (or) murein which gives stable form and mechanical strength to the cell. Peptidoglycogen consist of alternating units of N acetyl glucosamine and N-acetyl muramic acid with  $\beta$ -1,4 linkage. Gram positive bacteria have a much greater amount of peptidoglycan (50% or > of dry weight of bacterial cell wall) than gram negative bacteria (10% of dry weight of bacterial cell wall).

The peptidoglycan layer is again surrounded by another outer membrane that is rich in phospholipids proteins, lipopolysaccharides. This outer membrane is anchored to the underlying peptidoglycan layer by means of Braun's lipoprotein in case of gram negative bacteria. The gram positive bacteria have outer membrane that are rich with polysaccharides, teichoic acid and very little lipids or some time lipid is absent.

The electron microscopic studies of gram negative phytopathogenic bacteria clearly shows that outer most layer of cell wall is composed of protein and polysaccharides with whisker like extending out, second layer is chiefly made up of glycoproteins, third layer is possibly lipopolysaccharides, fourth may be protein and fifth inner most layer is a rigid peptidoglycan. From this it is clearly understood that peptidoglycan layer and any other boundary layer outside it constitute the cell wall (or) cell envelope. The cell wall appears to be separated from the cytoplasmic membrane by an electron transparent zone namely periplasmic space.

## 5. Cytoplasmic membrane

It is present immediately beneath the cell wall, usually 7.5 nm thick. It consists of 3 layers, the two outer layers are electron dense and about 2.5 nm thick. The middle layer is much less electron dense and about 5 nm thick. The cytoplasmic membrane is composed of protein (50%), lipids (25%) and carbohydrate (25%) and has three distinguished unique functions.

1. It behaves as an osmotic barrier so as to maintain favourable intracellular osmotic pressure.
2. Act as a house of permease (specific active transport system).
3. Act as a site for enzymatic reaction involved in energy metabolism.

These vital functions imply that these are denoted as an extremely important functional structure and any damage to it can result in death of the cell.

The cytoplasmic membrane penetrates into the cytoplasm and develops intracellular membranous structures. These are called as mesosome and are frequently observed in gram negative bacteria. Mesosome may be of two types.

1. Central
2. Peripheral

The former one is located near the middle of the cell. Due to the deep penetration of cytoplasmic membrane. It is involved in the DNA replication and cell multiplication. The latter one has shallow penetration into the cytoplasm and they seem to be involved in export of extracellular enzymes such as penicillinase.

## 6. Cytoplasm

It comprises of all the different substances and structures except nucleus inside the cell membrane. All particulate matter in the cytoplasm is surrounded by and suspended in an aqueous fluid or matrix.

## 7. Cytoplasmic inclusions

### i) Ribosome

Minutely granulated, diffused scattered macro molecular RNA protein bodies accumulated close to the cell membrane. They have the sedimentation co-efficient of 70s (swedsberg units).

### ii) Innert inclusions

Nothing but the storage bodies with food reserves such as lipids, polysaccharides, volutin etc.

### iii) Chromatophores

This may be present in photosynthetic cells and are self duplicating like chloroplast. Plant pathogenic bacteria does not have chromatophore.

### iv) Nucleus

Nucleus of the prokaryotic microorganism are more precisely called as nucleoid as if they neither have a distinguished closed nuclear membrane nor a mitotic apparatus.

Ris (1961) coined the term genophore to represent bacterial nucleus or chromosome. It occupies from one half to two-thirds of the intercellular space. Nucleus consist of a double stranded DNA fibrils. Genophore is a closed loop i.e. circular chromosome carries all the genetic information necessary to specify the essential properties of the prokaryotic cell. Histones, the base protein characteristics of eukaryotic cell are absent.

In case of *scheria E. coli*, DNA is a single molecule of 1100 - 1400  $\mu\text{m}$  in length. In addition to genophore, the bacterial cells also contain a small extra chromosomal circular DNA that is capable of autonomous multiplication called as plasmids. These plasmids also act as genetic determinants.

## **IV) Reproduction in bacteria**

Bacterial cell undergo reproduction both asexually and sexually. Rod shaped plant pathogenic bacteria reproduce by the asexual process known as binary fission or fission. In addition to this, bacterial cells also produce spores either within the cell (endospore) or external to the cell (exospore).

### **1. Binary fission**

By this process, the single cell of the bacterium split transversely into two equal cells. Inward growth of the cytoplasmic membrane towards the center of the cell, forming a transverse partition leads to the bifurcation of cytoplasm into approximately equal parts. Two layers of cell wall is synthesised between two layers of the cell membrane. When the cell wall formation is completed, two layers separate, splitting the two cells apart.

During the time, the DNA, which is in the form of a circular chromosome condenses into an amorphous mass, then elongates and becomes dumb-bell shaped before it divides into two equal pieces of daughter nuclei. During the division of DNA, plasmids also duplicate and distribute themselves equally in the two cell.

Under favourable condition, bacteria may divide every 20 - 50 minutes, one bacterium becoming two, two becoming four and so on. At this rate, 1 million progeny are produced in less than a day. Bacteria seldom maintain logarithmic growth for long period and growth is limited by exhaustion of available nutrients/accumulation of toxic metabolic waste products.

### **3. Endospore**

These are unique to bacteria and are rarely produced by plant pathogenic bacteria. Endospores are produced mostly by *Bacillus* and *Closteridium* singly one in each cell and are actually dormant cell, resistant to desiccation, staining, disinfecting chemicals, radiation and heat. These are usually produced after active growth and

multiplication Whenever there is nutrient deficiency. All endospores contain large amount of dipicolinic acid (DPA), a unique compound that is absent in the vegetative cells. During favourable condition, these dormant spore germinate and develop in to a new cell.

Endospore development involves four main stages.

1. Accumulation of cellular DNA to form axial filament and formation of transverse spore septum.
2. Auxial filament is then surrounded by cell membrane to form forespore.
3. Formation of cortex and other resistant materials around forespore, completing formation of spore, disintegration of remaining cell to liberate the free endospore.
4. Germination of endospore when condition become favourable.

The matured endospore have core in the centre which is surrounded in sequence by core membrane, core wall, cortex, inner coat layer, outer coat layer and exosporium basal layer.

In addition to this, bacteria also reproduce sexually by conjunction, transformtion and transduction.

## **1. Conjugation**

In this process, two cells come in contact among which one is called as donar and the other one is receptor cell. Usually the recipient cells have specific receptor site for conjucation and donar cells have special appendages called sex pili. The recipient cell is attacked to the tip of a sex pilus. Minute portion of donar DNA can passes through the sex plus into the recipient cell. The receptor cell after accepting the DNA divides by fission and the progeny thus contains a combination of genetic material of the two parents.

## **2. Transformation**

One bacterial cell absorbs the genetic material secreted (or) liberated by the rupture of cell wall of another bacterial cell. Here the compatibility is supposed to be due to the presence of a specific protein of the recipient cell.

## **3. Transduction**

It is just like transformation except that in the process, DNA from one cell is transferred into another by a virus (bacterophage) which infects the bacterium.

## **LECTURE NO.26**

### **Classification of bacteria**

Bacteria have some unique characteristics which distinguishes it. From the other living microorganism Haeckel (1866) who first proposed the kingdom Protista to include all unicellular, microscopic microorganism and placed various orgnaisms of Thallophyta (Plants) and protozoa (animals) in protesta. Later Chalton (1937) proposed prokaryotes and Cukaryotes based on general pattern of cellular organisation. Eucaryotic cell are generally characterised by the presence of membrane bound nucleus and membrane bound organelles such as mitochondria, chloroplast, golgi apparatus etc. in the cytoplasm while in prokaryotic cells, nucleus is not enclosed in a nuclear membrane and distributed as a discrete massess throughout the cytoplsm as well as there is no membrane bound organelle in the cytoplasm. In 1969, stainer considered prokaryoties as lower protist (Bacteria and blue green algae) and eukaryotes as higher protist (fungi, algae, protozoa). In the same year whittaker proposed a new kingdom Monera to keep the lower protist. After that the kingdom Prokaryotae was proposed for these protokaryotic microorganism by Allsopp.

The classification of bacteria is based mainly on morphology including flagellation, stain reaction, cultural, biochemical and physiological characters, serology, phage and bacteriocin typing, DNA base composition and homology, extensive

pathogenicity and host range tests with detailed description of symptom. The description should be published in the international journal of systematic bacteriology (IJSB) which is the official publication of international committee on systematic bacteriology.

The names of the bacteria are governed by the international code of nomenclature of bacteria. Many guidelines for describing bacteria are used in Bergey's manual of systematic bacteriology, the authenticated text on classification of all known bacteria. The latest edition (8th) of the manual includes all prokaryotic microorganism in the kingdom prokaryotae. Prokaryotae is distinguishable into two divisions (1) Bacteria and cyanobacteria. Bacteria are grouped under the class Schizomycetes, which contains 10 orders.

- |                        |                     |
|------------------------|---------------------|
| 1. Pseudomonadales     | 6. Bacteriales      |
| 2. Eubacteriales       | 7. Myxobacteriales  |
| 3. Chlamydobacteriales | 8. Spirochaetales   |
| 4. Hyphomicrobiales    | 9. Actinomycetales  |
| 5. Caryophanales       | 10. Mycoplasmatales |

Of which, Pseudomonadales and Eubacteriales contain organisms of rod, cocci and spiral forms.

I) A brief outline of classification of plant pathogenic bacteria based on shape are given below.

#### **IV) Salient features of important plant pathogenic genera**

The main characteristics of some of the most common plant pathogenic bacteria are discussed hereunder.

##### **1. Agrobacterium sp**

Gram negative, Rod shaped,  $0.8 \times 1.5 \times 3 \mu\text{m}$  size bacteria with 1-4 peritrichous flagella (or) single lateral flagella, produce abundant slime when grown on carbohydrate

containing media and produce non pigmented smooth colonies on culture media. These are best examples for soil inhabitant enters into the host mostly through wounds and caused crown gall disease. One important pathogenic species is *A. tumefaciens* (crown gall in apple) and *A. rhizopgenes* (hairy root of apple). The *A. tumefaciens* induces tumours/gall through T<sub>1</sub> plasmid (tumour inducing plasmid) which is a circular ds DNA molecule contain upto 2,00,000 base pairs organized into several give. T<sub>1</sub> plasmid contains TDNA region which induces the plants to synthesis IAA that is responsible for hypertrophy and hyperplasia of root tissue. T<sub>1</sub> plasmid is nowadays widely used as vectors in genetic engineering.

## **2) Corynebacterium (*Clavibacter/Curtobacter* sp)**

Gram positive, straight to slightly curved rod shape bacteria 0.5 - 0.9 x 1.5 - 4 µm in size sometimes produces stained segments or granules and club shaped swellings. Generally non motile but some species are motile by means of 1 or 2 polar flagella. Few sps have xylem limited growth some important species under this genera are *c. michiganense* bsp. *mechiganense* (Bacterial canker of tomato) *C. michiganense* subsp. *Sepidonicum* (ring spot of potato), *C. tritici* (yellow ear rot of whet), *C. flaccumfaciens* sub sp. *flaccumfaciens* (Bacterial wilt of beans). After 1980, these plant pathogenic sps under this were transferred to other genera such as *curtobacterium*, *clavibacter*, *Rodococcus*, *Arthrobactis* based on cell wall content, G and C content of DNA and genetic homologies. the abvoe five sps. have been transferred to the genera *clavibacter*. The *c. fascians*, that cause fascidation disase of many plant sps. is the only pathogen reclassified under the genus *Rodococcus* as *R. fascians*.

## **3. *Erwinia* sp. (*Pantoea* sp)**

Gram negative, straight rods of 0.5 - 1.00 x 1 - 3 µm in size, and are motile by several peritrichous flagella. *Erwinia* are the only plant pathogenic bacteia that are facultative anaerobes. Many speices of *Erwinia* have strong pectionolyte activities (*E.*

*carotovora*) and cause soft rot disease in vegetables but some sps of *Erwinia* does not produce pectic enzymes and cause necrotic wilt diseases in crop plants (*E. amylovora*). Important species are

1. *E. carotovora* pv. atroseptica Potato black kg,
2. *E.c.* sub sp. *carotovora* soft rot of vegetable
3. *E. amylovora* - (fire blight in apple and pear)
4. *E. stewartii* - (Corn wilt/Stewart's welt)
5. *E. tracheiphila* - (Cucumber welt)

#### **4. *Pseudomonas* sp. (*Ralstonia* sp/*Burkholderia* sp)**

These are gram negative, straight to curved rods of  $0.5 - 1.0 \times 15 - 4 \mu\text{M}$  size, motile by means of 1 (or) many polar flagella. Many sps are soil inhabitants or present in freshwater and even in marine. Most sps. of *Pseudomonas* infect plants and cause wilt, leafspot and gall symptom. Some plant pathogenic *Pseudomonas* produce yellow green diffusible fluorescent, pigments on artificial culture media and are called as fluorescent pseudomonad (eg) *Ps. syringae* while some sps do not produce any fluorescent pigments and are called as non-fluorescent pseudomonads. The various diseases produced by different sps of *Pseudomonas* are listed below.

1.	Ps syringae pv. phaseolicola	(Bean halo blight)
2.	Ps S. pv. lacrymans	(angular leaf spot of cucumber)
3.	Ps S. pv. tomato	(Bacterial speck of tomato)
4.	Ps S. pv tabaci	(Ps tabaci) - (Wild fire of tobacco)
5.	Ps andropogonis	(Bacterial stripe of sorghum and maize)
6.	Ps rubrilineans	(top rot of sugarcane/red stripe of sugarcane)
7.	Ps solanacearum	Bacterial wilt of solanaceous plants and moko wilt

		of banana
8.	Ps fluorescens	Pink eye disease of potato, slippery skin disease of onion and sour skin of onion
9.	Ps s. subsp savastanoi	Olive knot disease, bacterial gall/canker of oleander

## 5) Xanthomonas

Gram negative, straight rods of  $0.4 - 1.0 \times 1.2 - 3 \mu\text{m}$  size, move by means of polar flagella. Growth on agar media are usually yellow and grow very slowly and produce acid from glucose. It causes leaf spot, blight, cankers, leaf streak rots in crop plants. The important pathogenic sps under this genera are.

1. *X. oryzae* pv. *Oryzae* (Bacterial leaf blight in paddy)
2. *X.o.* pv. *oryzicola* (Bacterial leaf streak in paddy)
3. *X.c.* pv. *camppestris* (Black rot of crucifers)
4. *X.c.* pv. *malvacearum* (Black arm/angular leaf spot of cotton)
5. *X.c.* pv. *vesicatoria* (Bacterial leaf spot of tomato and chillies)
6. *X.c.* pv. *translucens* (bacterial blight of cereals)
7. *X.c.* pv. *vasculorum* (gummosis in sugarcane)
8. *X. albilineans* (sugarcane leaf scald)
9. *X. axonopodis* - (*X.c.* pv. *citre*) - Citrus canker.

## 6) Streptomyces

It is a gram positive bacter, produces slender, branched hyphae without cross walls,  $0.5 - 2 \mu\text{m}$  in diameter. At maturity, the aerial mycelium forms chains of 3 to many spores. On nutrient media, colonies are small (1 - 10 mm in diameter) at first with a smooth surface but later with a welt of aerial mycelium that may appear granular, powdery or velvety. They also produce antibiotics which act against another bacteria,

fungi and other microorganism. Most of them are soil inhabitants. The important species are.

1. *S. scabies* (Potato scab)
2. *S. ipomoea* (soil rot/pox of sweet potato)

## **LECTURE NO. 27**

### **SYMPTOMS OF BACTERIAL DISEASES**

Plant pathogenic bacteria are basically facultative saprophytes and they enter into the host plant mostly through wounds and natural openings and also through non cutinized areas like root hairs and stigma. They are unable to penetrate the cutinized tissues such as cuticle and periderm. Inside the host cell, the bacterium grow inter cellularly or intracellularly or intravascularly or combination of above means. at that time, the host tissue also react differently to the invading bacteria and exhibit different kinds of symptom.

Plant pathogenic bacteria causes a variety of symptoms like leaf spot, blight soft rot, canker, gall and tumours, wilt and scab symptoms in plant. The pathogen belonging to several genera causes same type of symptom and individual species a genus also capable causing different types of species.

The important symptoms are

#### **1. Leaf spot**

Symptom include the appearance of water soaked, circular or irregular, necrotic spot on leaves. Some time the spots are encircled with a spot on leaves. Some time the spots are encircled with a yellow halo.

In case of dicots, the development of such spot is restricted by intermediate or large veins and that spots become *angular*. However in monocots, the spots usually appears as *streaks* or *stripes*.

- (eg) Bacterial leaf spot in tomato : *X.c. pv. vesicatoria*  
 Angular leaf spot in cotton : *X.c. pv. malvacearum*  
 Bacterial leaf spot in pepper : *X.c. pv. betlicoal*  
 Bacterial leaf stripe in arecanut : *X.c. pv. arecae*  
 Bacterial leaf streak in paddy : *X.c. pv. oryzicola*

## 2. Leaf blight

Development of rapid and extensive necrosis of affected leaves resulting in scorched appearance.

- (eg) Bacterial blight in paddy : *X.c. pv. Oryzae*  
 Bacterial blight in beans : *X.c. pv. Phaseoli.*

## 3. Soft rot

Softening of tissues due to dissolution of middle lamella by enzyme and disintegration tissues and very often a dirty liquid oozes out of the affected part. Affected area is soft, watery, discoloured and somewhat depressed, or wrinkled or blistered. There well be a well defined demarkation between the sound and decayed tissues.

(eg) Soft rot of tomato, egg plant, crucifers, onion, carrot, celery, lettuce is caused by *Erwinia carotovora* pv. *carotovora*.

## 4. Canker

Formation of corky outgrowth on the surface of leaves, twigs and fruits as a result of necrosis of host tissues and reaction of undamaged tissues to produce corky cells. These cankerous growth are usually localized deep seated and confined to the parenchymatous tissues of the host plant. The cankers involve the cambium layer and entail the destruction of woody tissues in case of citrus, mango and many forest trees.

- (eg) Citrus canker : *X.c. pv. citre*  
 Bacterial canker in mango : *X.c. pv. mangiferae indica*

In citrus canker, symptoms, initially appear as small, watery, translucent spots of yellow colour which later mature to become white or greyish and finally rupture in the centre giving a rough, hard, corky an craker like appearance.

### **5. Scab**

The infected area become rough, corky, slightly raised with rusty surface and pitted due to the abnormal proliferation of tissues in the epidermis. When compared to canker, scab are also corky out growth but formed only by epidermal infection and are not deep seated.

(eg) Potato scab :*Streptomyces scabies*

### **6) Galls and tumours**

Development of glucose, elongated (or) irregular targe sized outgrowth on the affected plant part due to the hypertrophy and hyper plasma of cell. Hypertrophy means abnormal increase in size of individual cells and hyper plasia means abnormal increase in number of cells as a result of cell division (eg) crown gall in apple – *Agrobacterium tumefaciens*. This bacteria contains tumous inducing ( $T_1$ ) plasmid induce crown gall. This bacteria enters into the plants through wounds and stimulate the cells to divide and enlarge abnormally.

### **7. Wilt**

Yellowing, dropping, welting and death of the above ground parts of the plant.

In general, bacteria enter and multiply inside the xylem vessels of host plant. The occlusion of vessels by bacterial cells and their polysaccharides interfer with translocation of water and nutrients and Wilt symptom develops. Bacteria often destroys the cell wall of xylem vessels during disease development where as in case of fungal wilt, the cells of vascular bundles remain intact until the death of the plant.

(eg) Bacterial with in cucumber : *Erwinia tracheiphila*

Bacterial wilt in solanaceous plants : *Pseudomonas solanacearum*

The wilted plants show brown to black colour discolouration in vascular bundle. This may be due to the oxidation of phenolics to quinones by phenol oxidase enzyme

secreted by bacterial cells. This oxidized phenolics are inturn polymerized to form melorium which actually impart the colour to the vascular bundles.

### **Mechanism of action of bacterial pathogens**

Bacterial pathogens mostly enter into the host plant through noncuticularised areas such as root hairs, stigma and through some natural openings (Stomata, hydathods, lenticels) and mostly through wounds. The entered pathogens act on host plant in various ways and exhibit different kinds of symptom. The different mechanism of action of phytobacteria on host plant in the development of symptom are.

#### **1. Production of enzymes**

The soft rot bacteria (*Erwinia carotovora* subsp *Carotovora*) produces pectic enzymes like pectinoesterase and polygalacturonase extracellularly after they have entered into the host. as a result the cell wall are progressively destroyed before the bacterium enters into the cell cavity. In some bacteria the pectinolytic enzymes induces wilting.

#### **2. Production of toxin**

Some of the bacterial diseases are the direct effect of toxins either prouced by the bacterium or by bacteium host interaction.

- |                                |  |
|--------------------------------|--|
| (eg) Five blight in apple      | - <i>Erwinia amylovora</i> (Amylovorin)                  |
| Bacterial spot in fruits crops | - <i>Ps. Syringae</i> pv. <i>Syringae</i> (Syringamycen) |
| Wilt fire in tobacco           | - <i>Ps. tabaci</i> (Tabtoxin)                           |
| Halo blight of beans           | - <i>Ps.S. pv. Phaseolicola</i> (Phaseotoxin)            |

#### **3. Blocking of xylem vessel**

Many vascular wilt inducing bacteria grow inside the xylem vessel and plug the vessels which affect the water and nutrient transport. Beside thus, the enzymatic break down products of cell wall are also carried in the transpiration stream, collect at the vessel ends, form gel and gums that clog the vessel pores.

#### **4. Oxidation of phenol**

The oxidation of phenolics to quinones by phenoloxidases secreted by the bacterial cell in case of vascular wilt diseases.

## **5. Hypertrophy and hyperplasia**

The crown gall bacterium induces the host cell to undergo hypertrophy and hyperplasim and result in the development of tumours or galls. The tumour inducing principle (Ti) is located in the plasmid in the T DNA part. This T-DNA induces the cells to produce more IAA. Which is responsible for the excessive elongation and devision of cells. Once this plasmid get integrated with the hos DNA then the host cell become a tumour cell and there is no further role o the bacterium Tumour cell multiply and develop galls and this process cannot be checked by thalling the bacteria.

## **6. Production of polysaccharidis**

The bacterium *E. stewartii* produce more polyssaccharise which result in development of wilt.

### **Role of insects in dispersal**

Most of hopper nymphs can acquire phytoplasmas much readily or better than adult insects. The acquisition feeding period varies from several hours to days. The vectors transmit the pathogen only after an incubation period of 10-45 days. Mean while, phytoplasmas multiplies first in the alimentary canal than in to the hemolymph, brain and finally in the salivary glands of the insects. Once the concentration of phytoplasma reaches a certain level, then the vectors can able to transmit it into new plant more (or) less efficiently for the rest of its life. Though the phytoplasmas survive through subsequent molting, the insect must feed on infected plants in order to become more viruliferous. No transovarial transmission was not observed in any phytoplasma diseases.

The mode of transmission of some phytoplasmal diseases are given insects.

1. Peach : Leaf hopper and dodder (*Cuscuta*)  
*(Exitiances fusconervosus)*
2. Peach yellows (or) : Leaf hopper (*Micropsis trimaculata*) and by dodder  
Poja rosette
3. Pear decline : Psyllid (*Psylla pyricola*)

- 4. Mulberry dwarf : Leaf hopper (*Hishimonas sellatus*)
- 5. Tomato big bud : Leaf hopper (*Orosius argentatus*)
- 6. Tomato marginal : Leaf hopper (*Orosius albicinctus*)  
flavascence
- 7. Cucurbit phyllody : Leaf hopper (*O. albicinctus*)
- 8. Bitter gourd witches broom : Leaf hopper (*O. albicinctus*)
- 9. potato purple top roll : Leaf hopper (*O. albicinctus*)
- 10. Potato marginal flavascence : Leaf hopper (*O. albicinctus*)
- 11. Potato witches broom
- 12. Jasmine phyllody : White fly (*Dialeurodes kerkaldii*)
- 13. Anion aster yellows : Leaf hopper (*Macrosteles fasciform*)
- 14. Sugarcane white leaf : *Matsumuratellis hiroglyphicus*
- 15. Gingelly phyllody : Leaf hopper (*O. albicinctus*)
- 16. Brinjal little leaf : Leaf hopper (*Hishimonas phycitis*)
- 17. Sugarcane grassy shoot : Aphid (*Rhopalosiphum maidus*)
- 18. Coconut cadang – Cadang – Planthopper (*Myndus crudus*)
- 19. Sandal spike : *Moonia albimaculata and Jasus indicus*

### **Grafting**

- 1. Grapevine little leaf
- 2. Ber witches broom
- 3. Apple proliferation
- 4. Lab lab little leaf
- 5. Cucumber phyllody
- 6. Bitter gourd witches broom
- 7. Potato phyllody
- 8. Cotton stenosis/small leaf
- 9. Clove little leaf
- 10. Sunflower aster yellows and phyllody.

## LECTURE NO.28

### Plant virology

#### **Introduction**

Era of plant virology started in 1885 after invention of contagious nature of tobacco mosaic disease by the German scientist *Adolf Edward Mayer*. He first used the term mosaic (or) mosaikkrankheit for the appearance alternate yellow and green mottling in tobacco leaves. Later in 1890, IWA noski from Russia, observed this contagious agent is very small and it could pass through bacterial filter. In 1897, M.W. Beijerinck confirmed that the agent is not a bacterium but a contagium vivum fluidum. He is actually considered as founder of virology. He only used the term 'virus' for the contagious agent.

The perusal of the literatures shown that there are many scientist made significant contribution in the science of plant virology and it grows like a banian tree with many advances.

#### **Virus**

These are the submicroscopic, infectious obligate parasites, do not have metabolism of their own but depends upon other living cells for their multiplication. They neither have any sexual reproduction nor produce any resting structures as that of fungi and bacteria.

Today more than 500 plant viral diseases are known to cause heavy qualitative and quantitative reduction in yield. Virus could cause diseases in crop plants because of utilization of cellular substances during its multiplication inside the cells, taking up space in cells and finally due to disruption of cellular process. one virus can affect one or more different species of plants and even each plant species is affected by different kind of viruses. In India, every year, potato crop is severely affected by potato leaf roll virus and potato virus.

#### **Characters of virus**

1. It is a unique entity which is neither living nor non living but a macromolecule on the threshold of life. They behave like microorganism by causing disease, able to

replicate and have genetic functions but behave as a chemical molecule by crystallized into a powder and do not grow no metabolic enzymes.

2. They are not free living, purely obligate in nature, contain no metabolic enzymes (or) protein synthetic machinery of their own but they use the host machinery for their metabolic activities.

3. Virus must have nucleic acid and a protein coat. Nucleic acid carries the genome of the virus which differs from one virus to another. Nucleic acid must be either DNA or RNA and never both. It may be single stranded (or) double stranded and may be single segmented or with a few segments.

4. Protein coat present as a envelop around the nucleic acid throughout its active phase except at the time of replication when the protein coat and nucleic acid are separated.

5. They are neither cells nor consists of cells.

### **Morphology and structure**

Viruses are the intracellular obligate parasites which consist of nucleic acid and protein coat (capsid). Hence they may often called as nucleocapsid. The matured virus particle are called as virions which are infective in nature.

Virus are mostly elongated or spherical or variants of these two basic structures. The elongated virus may be rigid rods (15 - 30 nm x 480 - 2000 nm size) or flexible threads (10 - 13 nm x 480 - 2000 nm size). Similarly the spherical virus may be isometric or polyhedral (17 nm - 60 nm dia) in shape. Nearly half of the known viruses falls under category of elongated rods and admost many are spherical with the remaining being cylindrical bacillus like rods/rhabdoviruses.

### **Components of virus**

#### **a) Nucleic acid**

The virus particle contains only one kind of nucleic acid ie. DNA (or) RNA, which will serve as genetic apparatus. Since they are not cellular microorganism they do not have any separate nucleus, cytoplasm, other organelle etc.

Nucleic acid is strictly the infective component of the particle. Most of the plant virus consist of RNA but some virus also have DNA as its genetic component. Nucleic acid makes up 5 - 40% of the virus and the protein making up the remaining 60 - 95% of the virus particle. Elongated virus have low nucleic acid while spherical viruses contains high percentage of nucleic acid. The weight of nucleic acid ranges between 1 - 3 million ( $1-3 \times 10^6$ ).

Nucleic acid may be single stranded (SS) or double stranded (DS). Most plant virus (540) contains single strand RNA but 40 contains dsRNA, 30 contains dsDNA and about 50 contains ssDNA. In case of single strand nucleic acid, the strand may be free at both ends (linear nucleic acid strand) or the ends may be joined together to form a circle (circle nucleic acid strand). However, in double strand nucleic acid the two strands are coiled around each other helically.

The nucleic acid strand may be continuous (or) it may be present as two (or) more pieces in the same particle (or) different particle. Of the same protein subunit. When the genome is present in different particle the virus are known as multipartite virus or virus with a divided genome. In multipartite virus all the split genome present in different particle are essential for the successful infection and production of typical symptom. Some examples are give below

- 1) linear ss RNA in one piece.  
(eg) TMV, Tomato bushy stunt virus, Potato virus
- 2) Linear ssRNA in more than one piece contained in the same particle  
(eg) Turnip yellow virus, Potato leaf roll virus.
- 3) Linear ssRNA in more than one piece and each piece in a separate particle.  
(eg) Cucumber mosaic virus  
Cowpea mosaic virus.
- 4) Linear dsRNA in many piece but in one particle (eg) sugarcane fiji virus.
- 5) Circular ssDNA as in gemini virus  
(eg) Mungbean yellow mosaic virus  
Maize streak virus.

6) Circular dsDNA

(eg) Cauliflower mosaic virus.

In general nucleic acid consist of long chain like molecule consist of large number of nucleotides. Each nucleotide consist of a base, a five carbon sugar (either ribose in case of RNA) or deoxyribose in case of DNA) and phosphoric acid. The base may be adenine, guanine, thymine and cytosine in case of DNA and adenine, guanine, cytosine and uracil in case of RNA. The base is attached with the pentose sugar which in turn react with the phosphoric acid the sugar of one nucleotide react with phosphoric acid of another nucleotide and form long chains. The sequence of arrangement of nucleotide in the nucleic acid only specified the type of protein for the protein coat.

**b) Proteins : Coat**

The protein coat of the virus not only provides a protective sheathing for the nucleic acid of the virus but its presence generally increases of infectivity of nucleic acid. Like all proteins, viral protein is also made up of different amino acids which are linked by peptide bonds. The sequence of amino acid in the protein coat is dictated by the sequence of nucleotide in the nucleic acid.

Protein coat on capsid or shell is not smooth and it consist of a definite number of bumps which are called as protein subunits (or) structural units. Amino acid content for identical protein subunits are constant for a virus but vary for different viruses and even for different strains of the same virus. The clusters of structural unit (or) subunits are called as capsomeres.

The protein subunits are spirally arranged in elongated virus and are packed on the sides of polyhedral particles of the spherical viruses. In elongated virus the nucleic acid is arranged spirally where as in spherical virus, the nucleic acid is arranged in an unknown manner.

In some virus, there is also an envelope or mantle or limited membrane of lipid of lipoprotein outside the nucleoprotein such virus are called as lipoviruses. (eg) rhabdovirus like Potato yellow dwarf virus, tomato spotted wilt viruses, wheat striate mosaic virus.

In case of TMV, the protein sub unit consist of 158 aminoacid in a constant sequence and the protein subunits are united in a helix containing 16 1/2 subunits per turn. Each TMV particle consist of nearly 120 helix turn of protein subunits and the nucleic acid is parked tightly between the belives of protein subunits.

In some DNA viruses, two virus particles join and occur in twins. These virus are called as *Gemini virus*.

### **Satellite virus**

Satellite viruses are viruses which have small RNA genome and a protein coat. These virus must always be associated with another virus (*helper virus*) for its multiplication and plant infection. They usually acts as a parasite of the associated helper virus.

(eg) Tobacco necrosis Satellite virus.

### **Satellite RNA**

Satellite RNA have no coat protein of their own. These are small linear/circular RNA found inside virions of certain multicomponent virus. These multicomponent viruses helps in the multiplication satellite RNA and are called as helper virus.

The satellite RNA are not related/partially related to the RNA of helper virus and they may increase or decrease the infectivity of helper virus.

(eg) Satellite RNA in cucumber mosaic virus and tobacco ring spot virus.

In addition to this some time the satellite RNA are present in some satellite virus (eg) Tobacco necrosis virus contains small satellite RNA which depends upon tobacco necrosis virus for replication and on the satellite virus for encapsidation.

### **VIROID**

Virus without protein coat is called as viroid. They consist of only RNA and are subviral in size and reported to cause diseases to crop plants which well be dealt in detail in the succeeding chapter.

### **Virus infection and multiplication**

Virus enters into the plant cell through wounds made mechanically or by vectors or naturally by deposition into an ovule by an infected pollen grain.

Once the virus enters inside the host, it becomes naked by shedding its capsid or coat. In case of ssRNA virus, the naked RNA inside the host cell induces the cell to form the viral RNA polymerase. This RNA polymerase utilises the viral RNA strand as a template and forms complementary RNA (cRNA). The complementary RNA soon get separated from the original RNA strand and serve as a template for the synthesis of more viral RNA strand. These synthesised new viral strand or a segment of this act messenger RNA (mRNA) for the production of protein subunits. For this process also, virus utilises the aminoacids, ribosomes and tRNA of the host of the synthesis of its protein subunits. When the nucleic acid and virus protein subunits have been produced, the nucleic acid organics the protein subunits around it, assembled together and form the complete virus particle namely viroids. In case of RNA viruses (ssRNA or DS RNA) the entire process takes place in the cytoplasm of plant cell itself. Where as in ssDNA virus, the viroids are formed in the nucleus and then release into the cytoplasm of host cell. The first intact viroids appear in plant cell approximately 10 hours after inoculation. The multiplied virus particle existed with in the host cell as inclusion bodies.

The replication process varies slightly with different types of viral genome. In multipartite virus all the particles must be present in the same host cell for the replication and to develop infection. In some single strand negative RNA (-ss RNA), the RNA freed from the protein coat must be first transcribed into a positive strand RNA by a virus carried enzyme called transcriptase and then the remaining process of replication similar as mentioned above.

The process of replication in dsDNA is some what different. First the viral dsDNA enters into the cell nucleus and where it become twisted and supercoiled to form a minichromosome. This minichromosome like ds viral DNA is then transcribed into two single stood RNA's and transported into the cytoplasm where one small strand become translated into virus coat protein and the other large RNA strand become encapsidated by the coat protein subunits and is used as a template for the reverse transcriptase into a complete viroid ds DNA.

## **Movement of virus inside the plants**

Virus moves from one cell to another cell through the plasmodesmata, which connect the adjacent cells. Once the virus are injected into the phloem by some vectors then they are rapidly transported systemically over long distances within the plant through photosynthate stream and reenters the parenchyma cells adjacent to the phloem through plasmodesmata.

## **LECTURE NO.29**

### **Virus vector relationship**

(Based on length of temperature for which an insect remain viruliferous)

Persistence	Semipersistence (non circulative)	Non-persistence (non circulative)
Circulative	Non circulative	
Propagative	Non propagative	

Before going to see the each category in detail some important terminologies should be understood thoroughly.

### **1. Acquisition feeding period**

It is the time for which a virus free vector actually feeds on a virus infected plant to acquire the virus.

### **2. Acquisition access period**

It is the time for which a vector is allowed to feed on a source of virus.

### **3. Viruliferous vector**

Vector become able to transmit the virus. Generally, vector become viruliferous immediately after acquiring the virus in sufficient quantities from the diseased plant or there may be a waiting or latent period before the virus can be transmitted.

#### **4. Inoculation access period**

It is the time for which the vector after acquiring the virus is allowed to feed on a healthy plant to transmit virus.

#### **5. Inoculation feeding period**

The actual period of feeding to inoculate the virus is called as inoculation feeding period.

#### **6. Transfer time/transmission threshold period**

Minimum period of time that a virus needs for acquisition and subsequent transfer to a virus free plant is called as transfer time.

On the basis of length of time for which of insect vector remains viruliferous, they are classified as 1) Persistent (2) Semi persistent and (3) non persistent viruses.

##### **1) Persistent virus**

In case of persistent virus, the relationship is highly specific and the virus have an intimate biological relationship with the vector. Immediately after acquiring the virus the virus may be circulated through alimentary canal, gut wall and body fluid of the insect. Such virus have a latent period in vector body and persist for long in the vector. Molting has no effect on persistence such viruses are usually present in phloem of host cell and have long acquisition and inoculation feeding period. So they are also called as circulative virus sometimes the virus itself multiplies inside the vector body and they are called as circulative propagative virus. Certain viruses are carried/transferred to the progeny of vector such as eggs and are called as transovarial transmission. For example in paddy, the rice dwarf virus is transmitted to the egg masses of leaf hopper *Nephrotettix cineticeps*.

##### **2. Nonpersistent virus**

The acquisition and inoculation feeding period of such virus are usually few second and they do not have any latent period in the vector. Such virus are mechanically transmitted and are lost by the vector during molting.

### 3. Semipersistent virus

They have characters of both persistent and non persistent viruses. They have long acquisition feeding period to acquire virus from phloem of the host plant but the vector become immediately viruliferous without latency. Such virus persist in the vector of few days and are lost at motting. They are not circulated in the vector body.

**Vector :** any organism that carry diseases causing pathogen.

#### Difference between persistent and non-persistent virus

	Persistent virus	Non persistent virus
1.	The vectors retained the virus in their body throughout the life	The virus are retained only for a short period in the body of vector
2.	Viruses are retained even after moulting	Virus are lost form insect vector one the insect moults
3.	Acquisition feeding period is long (2 hrs)	Acquisition feeding period is short i.e. minimum of 15 sec. is enough
4.	There is latent period in vector	No latent period in the vector
5.	Long incubation period in the vector	Only short incubation period
6.	Virus can multiply in the vector body i.e. circulative and propagative	Do not multiply inside the vector

The insects involved in the viral transmission are dealt in detail hereunder.

#### i) Aphids

Aphids are the most important insect vectors known to transmit more than 250 plant viruses. Aphids may transmit the viruses in a persistent (circulative, non propagative and circulative propagative), semi persistent and non persistent way some time the non persistent way of transmission is called as stylet borne viruses. The viral pathogen that are transmitted through aphids are tested below.

Sl.No.	Virus	Vector	Type of transmission
1.	Bean common mosaic virus	<i>Aphis craccivora</i>	Non-persistent
2.	Citrus tristeza virus	<i>Toxoptera citricidus</i>	Non-persistent
3.	Potato virus Y	<i>Myzus persicae</i>	Non-persistent
4.	Barley yellow dwarf	<i>A. dirhodum</i>	Persistent
5.	Banana Bunchy top	<i>Pentolonia nigronervose</i>	Persistent
6.	Beet yellows	<i>M. Persicae</i>	Semi persistent
7.	Cardomum dwarf	<i>Micromyzus klimpongense</i>	Persistent
8.	Cardomum mosaic virus	<i>Pentalonia nigronervosa</i>	Non persistent
9.	Cauliflower mosaic virus	<i>Brevicorgne brassicae</i>	Semi persistent

### b) Leaf hoppers and plant hoppers

They rank second in transmitting when compare with aphids, they penetrate tissues more rapidly and cause more damage to cells virus. Most of the leaf hopper borne viruses are transmitted in a persistent manner (Circulatus and propagative) except few virus like Rice tungro virus of and maize chlorotic dwarf virus which is transmitted in a semi persistent way. Some of the examples of leaf hopper transmitted virus are.

1.	<i>Nephrotettix virescens</i>	Rice tungro virus
2.	<i>Cicadulinia cinai</i>	Maize streak virus
3.	<i>Nilaparvatha lugens</i>	Rice grassy stunt virus
4.	<i>Circulifer tenellus</i>	Beet curly top virus

5.	<i>Agallia constricta</i>	Potato yellow dwarf
6.	<i>Graminellis nigrifrons</i>	Maize chlorotic dwarf virus
7.	<i>Laodelphax striatellus</i>	Rice stripe virus
8.	<i>Nephrotettix cincticeps</i>	Rice dwarf
9.	<i>Sogatodes oryzicola</i>	Rice hoja blanca virus
10.	<i>Perkinsiella saccharida</i>	Sugarcane fiji virus

Among this, except rice tungro and maize chlorotic dwarf virus all are circulative and they require feeding period of one to several days before they become viruliferous but once days before they become viruliferous but once they have acquired virus they may remain viruliferous for the rest of their lives. transovarial type of transmission is noticed in rice dwarf virus.

### iii) White fly

Nymphs and adults of whiteflies transmit the viruses in a circulative and nonpropagative manner. Virus that are transmitted through white flies are economically important in tropical area and to a lesser extent in subtropical and temperate areas.

Many of the gemini viruses and some viruses belonging to earla virus, closterovirus, luteo virus, nepo virus and poty virus group are transmitted by white fly. Only three spp of whiteflies have been reported as vector of plant viruses of which *Bemisia tabaci* is the vector for many mosaic and leaf distortion symptom in infected plant. They usually need longer acquisition period than inoculation feeding period. The vectors mainly feed on phloem of the host plant and hence the virus are not sap transmissible. Some of the whitefly transmitted viruses are

Sl.No.	Virus	Vector
1.	Cassava mosaic virus	Bemisia tabaci
2.	Bhendi vein clearing virus	
3.	Cotton leaf curl, virus	
4.	Mungbean leaf curl virus	
5.	Tobacco leaf curl virus	
6.	Beet pseudo yellow virus	Trialeurodes vaporariorum

#### iv) Thrips

The unique characters of thrips as vector of plant virus is that only nymphs can acquire the virus and can remain viruliferous till death. They transmit the virus in a persistent and circulative manner and causes necrosis, bronzing and rosette type of symptom in plants. The species of thrips involved in virus transmission are *thrips tabaci*, *Thrips palmi*, *T. setosus*, *Frankliniella schultzei*, *F. fusca*, *F. occidentalis*, *Scirtothrips dorsalis*. The viruses transmitted by thrips include tomato spotted with virus (Tospo virus) tobacco ringspot virus (Nepo virus) and Tobacco streak virus (illar virus).

#### Mealy bug

Nymphs are more effective vectors than adults. Nineteen spp of mealy bug belonging to Pseudococcidae are reported to transmits six viruses in a semipersistent manner. Though they are phloem feeders, their efficacy in transmission of virus is comparatively less because they are not particularly mobile and rely on crawling to move from plant to plant. Some of the important mealy bud transmitted virus are listed below.

1.	Cocoa swollen shoot virus	Planococcoids njalensis
		Planococcus citri
		P. celtis, P. kenyae
		Pseudococcus longispinus
		Dysmicoccus brevipes
		Ferrisia virgala
2.	Pineapple wilt virus	Dysmicoccus brevipes

### Beetles

In contrast to aphids, thrips and whiteflies, beetles have chewing and biting mouthparts transmit four major group of viruses includes bromovirus, comovirus, tymovirus and sobemovirus. Usually the beetle transmitted viruses do not have no other vectors or only to a limited extent by others. The mode of transmission is persistent and circulative and there is not evidence for the propagation of virus inside the vector body. For example cowpea mosaic virus and southern bean mosaic virus are transmitted by *ceratoma trifurcata*. Squash mosaic virus by *Diabrotica longicomis* turnip yellow mosaic virus by *psylloides* sp: radish mosaic virus by *Epitrix hirtipennis*, Bhendi mosaic virus by *Podagria* sp, Broad bean stain virus by *Sitona lineata* and Bean pod mottle virus by *Epicauta vittata*.

### vi) Other insects

Other than these insects, some bugs, grasshoppers, earwigs, leaf miner are also reported to transmit some viral disease. They are

### Grasshopper

*Melanoplus differentialis* - Potato virus x Tobacco mosaic virus

### Earwig

*Foficula auricularia* - Turnip yellow mosaic

**Plant bug**

*Lygus protensis* - Spinach blight virus

**Lace bug**

*Piesma quadratum* - Beet leaf curl virus

**Lygaeid bug**

*Nysius* sp - Centrosima mosaic virus

**Leaf miner**

*Liriomyza langei* - Sowbane mosaic virus

Though the insects carry the viruses at great cost the virus may affect some vectors also to a limited extent. For example leaf hopper *delphacodes pellucida* carrying the wheat striate mosaic virus have lesser progeny and have a shorter life.

**g) Transmission by mites**

Among them, the eriophyid mites and spider mites have been shown to be vectors of plant viruses. Nine viruses are reported to be transmitted by seven different species of mites in a persistent circulative manner. The eriophid mites do not have much movement and are easily carried by wind because of their size (about 0.2 mm long and tiny). They have puncturing and sucking mouthparts. *Aceria cajani* transmits pigeonpea sterility mosaic virus, *A. tulipae* transmits wheat streak mosaic virus, *Abacarus hysterin* transmits ryegrass mosaic virus, *Eriophyes insidiosus* transmits peach mosaic virus and spider mites include *Brevipalpus obovatus* transmits citrus leprosis virus and *B. phoenicis* transmits coffee ring spot virus.

**h) Transmission by nematodes**

Soil inhabiting ectoparasitic nematodes transmit approximately 20 soil borne plant viruses whose infectivity will be lost when soil is allowed to dry at 20°C for a week. Raski and Goheen (1968) for the first time proved the association of *Xiphinema index* as the vector for the grapevine fan leaf virus. Four genera of nematode belonging

to Doriplaimoidea are known as vector of plant virus which are either polyhedral tubular.

The nematode transmitted viruses are broadly divided into two groups.

1. NEPO (Nematode transmitted polyhedral viruses)
2. NETU/TOBRA virus (Nematode transmitting tubular viruses)

Nepo viruses are vectored by *Xiphinema* sp, *Longidorus* sp and *Paralongidorus* sp. and NETU/TOBRA viruses are transmitted by species of *Trichodorus* and *Paratrichodorus*. Transmission of viruses by nematode resembles the transmission by insects NEPO virus are isometric with angular outlines of 23-30 nm. Whereas NETU viruses are straight tubular and rigid. Both NEPO and NETU are having bipartite genome with ssRNA, sap inoculated, seed (or) pollen transmitted and have moderate to wide host range *xiphinema* are transmitting the virus in a circulative manner while *Longidorus* transmits in a non persistent way. Regarding NETU/TOBRA virus some viruses are considered circulative whereas some of the strain of the same virus may be non persistent.

#### **Examples of nematode transmitted viruses :**

#### **NEPO viruses**

1.	<i>Xiphinema</i> index	Grafevine fan leaf virus
2.	<i>X. americanum</i>	Tomato ringspot virus, (Tom RV) Tobacco ringspot virus, (Tob RV) peach rosette mosaic virus (PRMV)
3.	<i>X. bakeri</i>	Arabis mosaic virus (AMV)
4.	<i>X. brevicolle</i>	Tomato ringspot virus (Tom RV)
5.	<i>X. coxi</i>	Cherry leaf roll virus (CLRV) Tobacco ringspot virus (Tom RV)

6.	<i>X. diversicandatum</i>	CLRV, GFV, Cowpea mosaic virus
7.	<i>X. italiae</i>	GFV
8.	<i>Rivesi</i>	Tom RV
9.	<i>Longidorus elongatus</i>	Tomato black ringspot virus (TBRV), CLRV
10.	<i>Longidorus macrosoma</i>	Raspberry ringspot virus
11.	<i>L. martini</i>	Prunus necrotic ringspot virus Mulberry ringspot virus
12.	<i>Paratongidorus maximus</i>	CLRV
	<b>NETU/TOBRA VIRUSES</b>	
1)	<i>Paratrichodorus allius</i>	Tobacco rattle virus (TRV)*
2)	<i>P. minor</i>	Tobacco rattle virus (TRV) Pepper ringspot virus
3)	<i>P. teres</i>	Pea early browning virus (PEBV)
4)	<i>P. Porosus</i>	TRV
5)	<i>Trichodorus cylindricus</i>	TRV
6)	<i>T. hopperi</i>	TRV
7)	<i>T. similis</i>	TRV
8)	<i>T. viruliferus and T. primitivus</i>	PEBV

\* TRV causes spraing disease in potato.

### i) Transmission through fungi

Fungi belong to the family plasmodiophoraceae, oomycetidae and synchytriaceae transmit soil borne viruses that survive for long period in dry soil condition.

These obligate fungi infect the host root by zoospores resting spore which carry the virus externally or internally and thus transmit the virus the persistence of virus depends on longevity of the resting spores of the fungi. *Olpidium brassicae* transmits tobacco necrosis virus, tobacco stunt virus lettuce big vein virus. *O. curcubilacearum* transmits cucumber necrosis virus. The species of *Olpidium* carries the virus particles only on the surface of zoospore and not on resting spores.

In contrast, species of *Polymyxa* and *spongospora* transmits the virus particles through their resting spores. The viruses are acquired during the colonization of virus infected plants and remain viable in the resting spores. Whenever these resting spores germinate, the virus is transmitted by zoospores into new roots. Some of the suitable examples under this category are (1) *Polymyxa graminis* (transmit Barley yellow mosaic, oat mosaic, wheat soil borne mosaic and wheat spindle streak mosaic) (2) *P. beta* (transmit Beet necrotic yellow vein or rhizomania of sugar beet) (3) *Spongospora subterranea* (transmits potato mop top).

### **Man**

Involvement of human being in virus transmission over short or long distance is highly significant as he did in fungi and bacterial dispersal. Majority of horticultural crops are being vegetatively propagated and he may be responsible for the transmission of virus during grafting, planting bulbs and corms cutting or by planting virus infected plants.

## **LECTURE NO.30**

### **Classification of plant viruses**

Scientist made several attempts to device a rational, taxonomically sound classification system. Originally the virus are recognised by the symptom they caused and are called based on symptom and name of the host (eg) Tobacco mosaic virus. In 1927, James Johnson proposed that the virus should be named only after the host and to

avoid the multiplicity of viruses on the same host, a distinguishing number should be given to each virus. Accordingly the first virus in tobacco is called as tobacco virus . Second as tobacco virus 2 and so on K.M. Smith proposed latin name like Nicotiana virus for tobacco virus in 1937. there was little bit confusion to understand the numerical codes by those people who were not working on the same crop so the letter codes were given such as Potato virus X, Y, S etc instead of numerical code. In 1939, F.O. Holmes proposed the Linnaeus system for virus nomenclature. Thus tobacco mosaic virus was called Marmor tabaci. In 1970, H.P. Hansen proposed another system based on fundamental characters of the virus. By this, tobacco mosaic virus is called as *Minchordia nicotianae* Here M means mechanically transmitted, chorda for rod shape and nicotiana for the host.

In 1966, Gibbs *et al.* classified the virus based on adansonian principles (grouping together of organisms showing greatest overall features are considered). The concentrational name of the virus is followed by a cryptogram.

Cryptogram consist of 4 pairs

1<sup>st</sup> pair : Type of nucleic acid (R = RNA; D = DNA)/Strandedness (1 = Single; 2 = double)

2<sup>nd</sup> pair : Molecular weight of nucleic acid in millions/Percentage of nucleic acid in infective particle.

3<sup>rd</sup> pair : Outline of particle/outline of nucleocapsid (nucleic acid + protein coat). The various symbols used are s = spherical; E= Elongated with parallel sides, ends not rounded; V = Elongated with parallel sides ends rounded x = complex.

3th pair : Kind of host infected/kind of vector. Symbols for host are a = algae; B – bacterium, F – Fungus; I – Invertibrate, M = Mycoplasma, P – Pteridophyte; S – Seed plant, V – vertiforate. Symbols for vectors are Ae –

Mite and tick; Al – White fly, Ap – Aphid, Au – Leaf/plant/tree hoppers; Cc – mealy bug Cl – beetle, Di – fly and mosquito; Fu – Fungus, Ne – nematode, Ps – psylla; th – thrips; Ve – Vector known but none of the above; O – spread without a vector.

Symbols for all pairs \* - Property of the virus not known.

( ) – Enclosed information doubtful/inconformed

[] – Enclosed cryptogram informs about a virus group.

For example the cryptogram of tobacco mosaic virus is R/1: 2/5: E/E: S/O.

Based on this cryptogram of important virus are listed below.

1. Tobamo virus	-	[R/1 : 2/5 : E/E : S/O]
2. Potex virus	-	[R/1 : 2.2/6 : E/E : S/O]
3. Poty virus	-	[R/1 : 3.5/5 : E/E : S/AP]
4. Curcumo virus	-	[R/1 : 1.3/9 : 0.8/19 : S/S: S/AP]
5. Caulimo virus	-	[D/2 : 4.5/16: S/S : S/AP]
6. Alfalfa mosaic virus	-	[R/1 : 1.1/16: 0.7/16 : U/U : S/AP]

## LECTURE NO.32

### Symptoms of phytoplasma diseases

They produce yellow type disease in plants. The important symptoms are gradual yellowing (or) reddening of leaves, reduction in size of leaves; shortening of internode; stunting, of plants; excessive proliferation of dormant buds into small branches which gives witches broom appearance; greening or sterility of flowers (Phyllody or antholysis or virescence); more or less dieback; decline and death of plants.

The yellowing of foliage, stunting and wilting of plants may be due to excessive callose formation and disintegration of phloem tissues in the infected plants.

Phytoplasmas also disrupt the growth regulators level in infected plants which lead to development of witches broom, phyllody and antholysis type of symptoms.

The symptom of some devastating phytoplasma diseases are explained here under.

### **1. Gingelly (sesame) phyllody**

Vein clearing, reduction in size of leaves, shortening of internode, stimulation of auxillary buds lead to profuse branching and conversion of floral parts into green leafy structures (Phyllody/virescence) are some of the characteristic symptoms. Diseased plants become completely sterile. This disease was transmitted with the help of leaf hopper, *orosius albicinetius*.

### **2. Brinjal (egg plant) little leaf**

Diseased plants produce small, narrow, thin, sessile, soft, glabrous and pale green colour leaves in a shortened internode. The plants look bushy due to the stimulation of auxillary buds to grow into shoot branches and are sterile. The leaf hopper, *Hishimonas phycitis* and *Empoasca devastans* transmit the from plant to plant.

### **3. Sugarcane grassy shoot**

The disease is otherwise called as new chlorotic disease or albinodisease or yellowing disease or bushy disease or leaf tuff.

Profuse tillering and grassy appearance of the shoot are the main symptoms. Leaves become narrow, yellow or entirely devoid of any pigments (albinism or white leaf), small like grass leaves and canes re then with short internode gives a bushy or grassy appearance to the clump. The vector responsible for the transmission is aphid, *Rhopalosiphum maidis* and *R. sacchari*.

### **4. Coconut lethal yellowing**

The first symptom is the dropping of prematurated coconuts of any size followed by blackening of inflorescence tip and all male flowers. Then the lower leaves exhibit yellowing which progress upward to the young one. later lower leaves die prematurely, turn brown and cling to the tree while the young one are becoming yellow. Finally all

the leaves and vegetative buds die, fall way and left only the trunk which looks like a telephone pole. This disease is transmitted by plant hoppers, *Myndus crudus*.

### **5. Sandal spike**

This disease was first reported from Coorg district of Karnataka state in India. The diseased plants exhibit 2 kinds of symptom.

#### **i. Rosette spike**

Symptoms include the reduction of leaf size, shortening of internode and attenuation of branches into stiff pointed structures resembling like spike either in isolated branches or in the whole tree. Later the leaves become yellowish and finally reddish brown before the death of the plant.

#### **ii) Pendulous spike**

It is quite rare. Here in this type, the auxillary buds remain dormant and the individual infected shoots show continuous apical growth and assume a dropping habit.

This disease is transmitted by grafting, dodder and also by insect vectors *Moonia albimaculata*, *Jasus indicus* and *Nephrotettix virescens*.

## **SPIROPLASMAS**

These are the helical mollicutes lacking a rigid cell wall but the cells are bounded by unit membrane of 8-10  $\mu\text{m}$  thickness, and are usually facultative saprophyte. Similar to bacteria, they have ribosome (RNA) and DNA in the prokaryotic nucleus. Unlike phytoplasmas, they can be cultured on artificial media, colonies on agar have 0-2 mm diameter and appears as granules but some have fried – egg appearance.

Spiroplasma are very in shape from spherical to slightly ovoid to helical and branched non helical structures. They produce helical forms in liquid culture media. They do not have any flagella but have rotary motility and reproduce mainly by fission.

Spiroplasmas are resistant to penicillin but inhibited by tetracycline and are transmitted mostly by leaf hoppers. The well known species found in many decot plant is *Spiroplasma citri*. The *S. citri* and other spiroplasmas also infect their respective insect vectors.

Citrus stub bom, horse radish brittle root disease, corn stunt and rice yellow dwarf are some diseases caused by spiroplasmas.

### **Citrus stub born**

The important symptoms are production of bunchy upright growth of twigs and branches with short internodes chlorosis of foliage and excessive number of shoots. Diseased plants exhibit excessive winter defoliation and produce small, lopsided fruits with thin rind from fruit equator to the stylar end fruits are usually sour and have an unpleasant odouo.

The causative agent is *Spiroplasma citri* a gram positive wall-less. It is found in the phloem and transmitted frequently by buding and grafting. In citrus orchards, leaf hoppers such as *Circulifer tenellus scaphytopius nitridus* and *Neoaliturus haemoceps* aids in the dispersal.

### **2. Corn stunt disease**

Infected plants are stunted, bushy with chlorotic leaves, produces numerous small ear with little or no seed and tassels are usually sterile.

Corn stunt steroplasma is transmitted in nature by the leaf hoppers *Dalbalus elimatus* and *D. maidisi*

### **3. Rice yellow dwarf**

Affected plants become dwarf with pale green or yellow narrow leaves and are sterile. The causative agent is transmitted mostly by green leaf bug *Trigopotylus ruficornis*.

### **L form bacteria**

'L' from bacteria are morphologically indistinguishable from the phytoplasm obsered in plants. True bacteria often produces variants which failed to produce cell walls. Such wall less bacteria are called as L-form (or) L-phase bacteria cell wall production. They are reverted to the original bacterial form when the substance inhibiting bacterial cell wall formation is removed from the culture media but some of them stable and are unable to revert to the original bacterium. However, L-form become able to induce disease without riversion to the bacterial parents. They are more

permeable and are sensitive to antibiotics that affect other cell functions beside cell wall synthesis.

Plant pathogenic bacteria such as *Agrobacterium tumefaciens* (crown gall disease) and *Erwinia carotovora* pv. *atroseptica* (black leg of potato) are reported to produce L-form bacteria. These organisms have pathogenicity similar to parent bacteria and could be reisolated and cultured from the infected portions.

## **VIROID**

Viroids are naked nucleic acid without coat protein. It is a smallest known agents of infectious disease (minivirus) with low molecular weight ribonucleic acid (RNA) that can infect plant cells, replicate themselves and cause disease. When compared with virus, viroid have 250-370 nucleotides in their RNA compared to virus which contain 4-2000 nucleotides. Then the viral RNA is enclosed in a protein coat while the viroid lack a protein coat. Only because of the small size (9250 – 370 nucleotide) RNA, viroid are not able to synthesis protein and are replicase enzyme required to replicate and solely rely on host plant for multiplication.

Viroids are circular, single stranded RNA molecules with excessive base pairing in parts of the RNA strand. The base pairing result in the formation of hairpin structure with single and double stranded regions of the same viroid and contributes to the stability of the RNA.

They seems to be associated with the cell nuclei, particularly the chromatin and possibly with the endomembrane system of the cell. They replicate by direct RNA copying in which all components required for viroid multiplication including RNA Polymerases are provided by the host. During replication, the circular (+) RNA strand acts as a rolling drum producing many copies of linear strand (-) RNA. These linear (-) RNA strands serve as a template for replication of multimeric strands of (+) RNA. This (+) RNA are subsequently cleaved by enzyme that release linear, unit length viroid (+) RNAs. Which circularise and produce many copies of the original viroid RNA.

Viroids are transmitted from the diseased to healthy plants by mechanical means through sap carried on hand (or) tools during propagation or cultural practices and most by vegetative propagation. No specific insect vectors are known. Viroid diseases such as potato spindle tuber, coconut cadang – cadang, tomato bunchy top and apple scar skin are reported to transmitted through pollen and seeds.

Viroids over winter or perrenate in perennial hosts which includes main host of almost all known viroids and also outside the host or in dead plant matter for few minutes to few months. The heat treatment of infected plants should not inactivate the viroids since they are quite resistant to high temperature.

The important viroid diseases are (1) coconut cadang – Cadang (2) citrus exocortis (3) Chrysanthemum stunt (4) Chrysanthemum chlorotic mottle (5) Potato spindle tuber (6) hop stunt and (7) cucumber pale fruit. These viroid diseases exhibit a variety of symptoms that resemble those caused by viral infection.

### **Potato spindle tuber (PSTVd)**

This is the first recognized viroid by Diener (1971). Infected plants appear erect, spindly and dwarfed. The leaves are small and erect the leaflets are dark green and some time show rolling and twisting. The diseased plants produce elongated tubers with tapering end. Such tubers are smoother but they have more numerous and conspicuous tuber eyes.

It is transmitted mechanically by culting knives. The during cultural practices and during handling are planting of crop. They are also seems to be transmitted by pollen, seed and by contaminated mouthparts of grass hoppers, flea beetles and bugs.

### **Citrus exocortis (CEVD)**

Diseased plants develop narrow vertical thin strips of partially loose outer bark that give the bark a cracked and scaly appearance such plants also exhibit yellow blotches on young infected stem and some citron plants show leaf and stem epinasty along with cracking and darkening of lef veins and petioles.

The citrus exocortis viroid (CEVd) is readily transmitted from diseased to healthy plants by budding knives, pruning shears, cutting tools, by hand and possibly by scratching and gnawing of animals.

### **3. Coconut cardang – Cadang (CCVd)**

CCVd is the smallest viroid known.

Coconut trees become infected with cadang-cadang after they have begun to flower. Diseased plants produce leaves with bright yellow spots and nuts become rounded and develop scarification on their surface. Two to four years later, the inflorescence are killed and as a result no more coconuts are produced. Five to seven years from the beginning of symptoms, the constantly increasing number of chlorotic spots gives the whole crown a yellowish or bronze colour, the number and size of fronds in the crown continue to be reduced, growing bud dies, fall off and finally left only the trunk which stands like a telephone pole.

This viroid is transmitted on the mouth parts of different insects, mechanically on the machetes used to cut steps at the base of the palm, to dislodge the cuts and to make cuts to the inflorescence for tapping their sugary sap and possibly through infected pollen.

Differences between the symptoms produced by virus infection and deficiency symptoms.

VIRUS	DEFICIENCY
Mosaic, streaks type symptoms are seen	Complete yellowing of leaves
The symptoms are seen throughout the field	Not seen at the edges of the field where N content is more
The symptoms may be temporarily recovered when micro nutrients are applied	The symptoms will be permanently recovered when the deficient micronutrient is applied.
The virus is vector or sap transmissible	It is not vector or sap transmissible
The virus responds to the serological tests such as Precipitation tests, ELISA etc.	This symptom does not respond to the serological tests.

## LECTURE 33

### ALGAE

Algae are the eukaryotic thallophytes have chlorophyll a as their primary photosynthetic pigments. These ubiquitous organisms have aquatic, terrestrial or subterranean habitat with a relatively undifferentiated plant body made up of single or many cells. But they lack vascular tissue. The study of algae (Singular alga) is known as phycology of algology.

Algal cells may be spherical, rod shaped, barrel shaped, club shaped or spindle shaped. The cell wall is thick and somewhat rigid, contain a well defined nucleus, chloroplast (contain photosynthetic pigments) mitochondria, vacuoles, oil droplets, starch grains etc. The cell wall is encircled by gelatinous matrix which was secreted through the cell wall. Some algae move by flagella and some other do not have any locomotion. But in some members only the zoospores (asexual reproductive cells) are motile.

#### **Classification of algae**

Algae belonging to the phylum chlorophyta are generally classified based on the following characters.

- Morphology and characteristic of cell and flagella
- Method of reproduction
- Nature and properties of photosynthetic pigments, stored product or assimilatory product.

The phylum chlorophyta have 7 subdivisions.

1. Chlorophycophyta (green algae)
2. Chrysophycophyta (Golden and yellow green algae including diatoms)
3. Cryptophycophyta (Crypto monads)
4. Euglenophycophyta (Euglenoids)
5. Phaeophycophyta (Brown algae)
6. Pyrrophytcophyta (Dinoflagellates)
7. Rhodophycophyta (Red algae)

Among the 7 sub divisions, the sub division chlorophycophyta are of more important in the view of plant pathologist sine the gram algae genera *Cephaleuros* infect many horticultural crops and reduces their economic value. The *Cephaleuros* causes '**Red Rust**' disease in crop plants. Suitable examples are the *C. parasiticus* infect tea, mango and citrus; *C. coffea* infect coffee; *C. mimus* infect cocoa; *C. virescens* infect tea.

### **Symptoms of red rust (*Cephaleuros* sp.)**

- Symptoms are noticed on leaves, stem and fruits
- Circular to irregular, yellowish green to orange to grey colour, velvety, raised, hairy pestulus mostly appear on the upper surface of the leaves. The rusty colour of the pustule may be due to presence of orange colour pigment (*haematochrome*) in the sporophore and sterile hairs. Once the spore get mature, fall off during favourable condition and they left cream to white velvety texture on the surface of the leaves. Affected leaves defoliate prematurely.
- Very rarely the pustules appear on the lower surface of the leaves. The pustules on stem, are elongated and lea to the formation of scaly and cracked bark.
- Affected plants are stunted and produce few chlorotic leaves
- Pustules on the fruits are initially dark grey but later turn brown and which guess ugly appearance to the fruits.

### **Characters of *Cephaleuros* sp.**

*Cephaleuros* sp produces disc like thallus comprises of symmetrically arranged barrel shaped cells which grow dichotomously from the centre to the periphery of the thallus. The centre of the thallus is multilayered while the periphery is single layered. The cells have chlorophyll a, b and carotenoid as photosynthetic pigments and cell wall contains cellulose, xylan mannan etc.

Usually it grows between the cuticle and epidermis of the host tissue and very rarely between epidermis and palisade cells. The host cell below this assume a corky appearance and prevent further penetration of algae. Rhizoids arise from the thallus extend to the underlying cells (cuticle and epidermis) by the mechanical forces of

thallus and not by any enzymatic dissolution of host surface. The rhizoids act as anchoring as well as absorbing organ and get nourishment by osmosis.

### **Reproduction**

It undergoes both sexual and asexual reproduction. The asexual progeny are more responsible for the quick spread and outbreak of diseases during favourable condition.

#### **Asexual reproduction**

Algae produces many sterile hairs (Setae) among the sporangiophore (sporophore) on the periphery of the thallus after rupturing the cuticle or epidermis. The sterile hairs and sporangiophore have orange colour pigments. The sterile hairs are pointed while the sporangiophore bulge at the tip to form a vesicle. Light orange colour protrusions or sterigmata come out from the vesicle which are usually four in number but sometime it may be eight. Each protrusions bear orange colour, oval to globose zoosporangium at the tip. The zoosporangia are disseminated by wind and released their zoospore (8-30) through the ostiole under suitable climatic conditions. Whenever this zoospore reaches host cell, it comes to rest, and cross walls are formed repeatedly. Bus develop from the undersurface penetrate the host cell and to develop new thallus.

#### **Sexual reproduction**

Algae undergoes sexual reproduction by isogametic copulation. Large, sessile, flask shaped gametangia are produced in the thallus. In the presence of water 8-32 biflagellate zoospores are released from each gametangium. These swarmspores act as gametes. Two gamete from the same or different gametangia fuse together to form zygote. The zygote develop into a dwarf sporophyte consisting of an attaching stalk bearing one or ore cells. Each cell has a small microsporangium. Four quadri flagellate micro zoospore are formed in each microporangium as result of one meiosis. These microzoospores very rarely infect the host tissues.

## LECTURE NO.34

### PHANEROGAMIC PARASITES DISEASES

Certain flowering plants (Phanerogams) also parasitize the crop plants in addition to the microorganisms. They mostly belong to Loranthaceae, Convolvulaceae, Scrophulariaceae, Orabanchaceae, Lauraceae, Santalaceae and Balauphoraceae. They produce flowers and seeds and parasitize their host by drawing nutrition and water. Some phanerogams have green leaves, roots and they have the ability to synthesis food materials but they obtain only the mineral constituents of food from the host, then they are called hemiparasite/waterparasite/partial parasite. Some of the phanerogams which do not have any chlorophyll completely depend on host for water and all minerals. They are called as holoparasite or complete or total parasite.

The phanerogamic plants are divided into.

1. Stem parasite

Total parasite --- Cuscuta

Semiparasite Loranthus

2. Root parasite

Total parasite	-	Orabanche
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Semiparasite	-	Striga
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Phanerogams have haustoria as absorbing organ, which are sent deep into the vascular bundle of the host to draw water and nutrients. The haustoria in general secrete some pectolytic and cellulolytic enzymes which soften the host tissue. Haustoria have higher osmotic pressure than that of host tissue which facilitate easy absorption of nutrients. The affected plants show stunting, chlorosis and death.

Based on the habit and attachment of the parasite with the host, they are commonly grouped as stem parasite and root parasite. They are again divided into total or semi parasite depending upon their mode of parasitism.

### a) Stem parasite

#### i) Complete/holo/total parasite: *Cuscuta* sp.

##### ***C. campestris, C. trifoli, C. planiflora Dodder C. indicora***

Commonly known as gold thread, hellvine, hair weed, devils hair and love vine.

- Attacks alfalfa, clover, onion, flax, sugar beet, potato, chillies many ornamentals etc.
- It is a yellow or orange vine strands which grow and twin the plant. They do not have leaves but bear only very minute scale leaves. Dodder produces flowers and fruits. Flowers are white, pink or yellowish, which form seed.
- On severe infection, they form a dense and tangled mat on the crop.
- Seeds of dodder overwinter in the infested soil, germinate to produce a slender yellow shoot, make contact with the susceptible host plant, encircle and send haustoria into the vascular bundle of the host.
- It does not produce any roots. As soon as the dodder is established with the host, base of the dodder shrivels, dries and cut off from the ground. Thus it completely depends upon the host for nutrients and water.
- Thus the affected plants get weakened and yield poorly.
- Seeds of cuscuta are mainly spread by animals, water and implements.

#### ii) Partial semi / hemi stem parasite

Commonly known Loranthus ,Giant mistletoe, Banda.

##### *Dendrophthae flacata*

(Order: Santalales; Family: Loranthaceae)

- Attacks mango, citrus, apple, rubber, guava etc.
- Partial parasite of tree trunks and branches with brown stem, dark green leaves but no roots.
- Stem of the parasite is usually thick, and flattened at the node, appears in clusters at the point of attack which can be easily spotted on the trees.
- At the point of attachment with the tree, it shows swellings or tumourous growth where the haustoria are produced.

- This parasite produces flowers which are long, tubular, greenish white or red and borne in clusters.
- It produces fleshy fruits with single seed. The affected host plant become stunted in growth with few small chlorotic leaves.
- Dispersal of the seed is mostly through the birds and to some extend by animals.

**b) Root parasite / total/holo/complete parasite (eg)**

- Commonly known as Broom rape or *Tokra*.
- *O. cernua* var. *dessertorum*, *O. robancro ramosa*, *O. minor*, *O. crenata*
- (*Order. Orchidales, Family. Orobanchaceae*)
- It is a serious parasite in tobacco, tomato, brinjal, cabbage, cauliflower etc.
- It is an annual fleshy flowering plant growing to a height of about 10 - 15" with pale cylindrical stem, thickened at base and covered with brown scaly leaves that end in spikes.
- Plants lack chlorophyll, flowers arise from axils of the scale leaves.
- Flowers have well developed lobed calyx, tubular corolla, superior ovary, numerous ovules and large four lobed stigma. Fruits are capsules containing small black reticulate and ovoid seed.
- Seeds remain dormant in the soil for many years and they germinate due to a stimulant (benzopyran derivatives) present in the root exudate of susceptible host plant Ethylene, gibberellin and coumarins also induce the seed germination.
- In tobacco it appears in clusters of 50 - 100 shoots around the base of a single plant 5 - 6 weeks after transplanting. Affected tobacco plants are stunted, show withering and drooping of leaves leading to wilting.

**ii) Hemi/partial/semi parasite.**

Commonly known as witch weed or striga.

*S. asiatica* parasitise sorghum, maize and sugarcane

*S. densiflora* parasitise sorghum and sugarcane.

- Mostly affect the monocots
- It is a small plant with bright green leaves grows upto a height of 15 - 30 cm.

- It occurs in clusters of 10 - 20/host plant.
- *S. asiastica* produces pink flowers while *S. densiflora* produces white flower with a pronounced bend in corolla tube.
- This phanerogam lack typical root hairs and root cap.
- Fruits contain minute seeds in abundance which survive in soil for many years.
- Seeds germinate after post harvest ripening of about two weeks, in response to the host stimulant *viz.*, *strigol* ethylene, cytokinin, gibberellin and coumarin in strigol.
- This parasite slowly attach to the host root by haustoria, grow below the soil surface and produce underground stem and roots for about 1-2 months. Then it grows faster and appears at the base of the host plant.
- Severe infection of striga causes yellowing and wilting of host leaves. Sometime the host plant may die.

### **Deficiency diseases / Nutritional disorders**

Nitrogen, phosphorus, potash, calcium, sulphur, magnesium are required comparatively in large amounts to the crop, hence they are called as major elements while ferous, boron, manganese, zinc, copper, chloride and molybdenum are required in very small amount, so they are called as micro/minor/trace elements .They develop hunger signs in the crop plants. Such symptoms are called as non-parasitic diseases/physiogenic diseases/physiological disorders/nutritional disorders /abiotic diseases.

#### **1. Nitrogen**

This nutrient is essential for chlorophyll proteins, enzymes and for all other compounds. In case of deficiency, plant growth is reduced, leaves become yellow or light brown, stem become slender and short (eg) Red leaf of cotton, Chlorosis in rice.

#### **2. Phosphorous**

It is a constituent of phospholipids, nucleic acid and many proteins.

Dark green leaves of deficient plants (lower leaves) become red to purple owing to the abnormal production of anthocyanin pigments. Sometimes, necrotic spots are noticed in the leaves and leaf margin showing scorching.

(eg) P. deficiency in sunflower, soyabean, peach.

### **3. Potash**

It is important for carbohydrate and protein synthesis. Act as a catalyst for many reaction.

Dull bluish colour leaves, tip burn, marginal scorching, brown spalling, rolling/curling of lamina, poor root growth and shortening of internode are observed.

(eg) Marginal drying in banana.

### **4. Calcium**

Involves in regulation of permeability of membrane, activity of many enzymes. Affected plants have uneven leaf growth, lamina shows scorching,(brown) marginal chlorosis, killing of growing tip and leaf tissues (eg) Blossom end rot of tomato.

### **5. Magnesium**

It is the structural element of certain enzymes of CHO synthesis. It act as cofactor for certain enzymes. Leaves show chlorosis in the form of interveinal mottling, Midrib remain green and gives inverted 'V' shaped green portion near the base of leves.

(eg) Mg deficiency in Tomato, potato.

### **6. Sulphur**

Certain vitamins, coenzymes and amino acids contain sulphur.

Younger leaves show yellowing, roll upward and are brittle in nature (eg) S' deficiency in coconut, cotton, citrus.

### **7. Iron**

Many respiratory enzymes have iron. In chrlorophyll synthesis, the element seems to play a catalytic role. Deficient plants bear leaves which become chlorotic with main veins remaining green.

(eg) chlorosis in sugarcane, grundnut, green netting of citrus.

### **8. Zinc**

Component of many enzymes of auxin synthesis and CHO oxidation.

Deficient plants show interveinal chlorosis followed by necrosis. Affected plants have shorter internode and small leaves, poor fruit, seed setting. The citrus leaves exhibit inverted ‘V’ shaped symptoms

(eg) Khaira disease of rice folia cellosis of citrus.

Citrus franching

### **9. Boron**

Deficient plants are thicker and brittle, tender growing point die, root are thick and stunted, internode shortened, storage tissues may show cracks/rot in the central part.

(eg)

### **10. Manganese**

Co factor of several enzymes of cellular respiration, N metabolism and photosynthesis.

Leaves of deficient plants show chlorosis, smallest vein remain green, finally chlorotic area become necrotic.

(eg) Mottle leaf in citrus

Pahala disease of sugarcane

### **11. Copper**

Co-factor for several oxidative enzymes. Deficient plants bears marginal chlorosis and withered tip. Die-back symptoms also noticed. In case of cereals, head become dwarfed and distorted.

(eg) exanthema in citrus

Reclamation disease of oat

### **12. Molybdenum**

In involved in the reduction of nitrate to nitrate. Deficient plants bear mottled and necrotic leaves with thiner and dry leaf lamina. Distortion and death of growing tissue may also take place (eg) Whip tail of cauliflower /cabbage.



