Dr. Sayantani Das, Assistant Professor
Department of Botany, Netaji Nagar Day College

E-CONTENT
Prepared for UG (Botany Hons. Courses under CBCS)

Course Name	Course Code	Credits	System	Syllabus
CORE COURSE- 2 MYCOLOGY AND PHYTO-PATHOLOGY	BOT-A-CC-1-2-TH	THEORETICAL (Credits 4, Lectures 60)	CBCS	University of Calcutta

CORE COURSE 2

MYCOLOGY AND PHYTO-PATHOLOGY (BOT-A-CC-1-2-TH)

THEORETICAL (Credits 4, Lectures 60)

PHYTO-PATHOLOGY

1. Terms and Definitions : 1.1. Disease concept, 1.2. Symptoms, 1.3. Etiology & causal complex, 1.4. Primary and secondary inocula, 1.5. Infection, 1.6. Pathogenecity and pathogenesis, 1.7. Necrotroph and Biotroph, 1.8. Koch's Postulates, 1.9. Endemic, Epidemic, Pandemic and Sporadic disease, 1.10. Disease triangle, 1.11. Disease cycle (monocyclic, polycyclic and polyetic).

2. Host – Parasite Interaction: 2.1. Mechanism of infection (Brief idea about Pre-penetration, Penetration and Post-penetration), 2.2. Pathotoxin (Definition,criteria and example), 2.3. Defense mechanism with special reference to Phytoalexin, 2.4. Resistance- Systemic acquired and Induced systemic. lectures

3. Plant Disease Management : 3.1. Quarantine, 3.2. Chemical, 3.3. Biological, 3.4. Integrated. l

4. Symptoms , Causal organism, Disease cycle and Control measures of: 4.1. Late blight of Potato, 4.2. Brown spot of rice, 4.3. Black stem rust of wheat, 4.4. Stem rot of jute. …….10 lecture lectures 4. Symptoms , Causal organism, Disease cycle and Control measures of: 4.1. Late blight of Potato, 4.2. Brown spot of rice, 4.3. Black stem rust of wheat, 4.4. Stem rot of jute.

Mechanism of Infection

A plant becomes diseased when it is attacked by a pathogen or affected by an abiotic agent. Therefore, for a plant disease to occur, atleast 2 components, plant and pathogen must come in contact with one another and must interact. Environmental conditions also play an important role because some virulent pathogens become avirulent at extreme temperature. Thus together the 3 components, susceptible host, an aggressive pathogen and a favourable environment together constitute a disease triangle, which bring about a disease. Recently another factor, time has been introduced. Thus the new concept nowadays is of the disease pyramid. Reducing any component of disease pyramid through management would decrease the degree of the disease in a plant or plant population. The disease develops and establishes itself through a chain of events which is called **pathogenesis,** whereasthe ability ofapathogen to cause the disease is its **pathogenicity**.

The chain of events leading to pathogenesis occurs in succession and together constitutes the disease cycle. The primary events in a disease cycle are

i. Inoculation/Contact

ii. Pre-penetration

iii. Penetration

iv. Establishment and colonisation

i. Inoculation/Contact:

Inoculation is the coming in contact of the pathogen with the host. The part of the pathogen that on coming in contact with the plant initiates disease is called inoculum. Thus in fungi, inocula may be spores, sclerotia or mycelial fragments. These are wind borne or water borne. Some fungi, bacteria and most of the viruses are carried to their host plants by insects, nematodes and other vectors. Motile propagules of fungal pathogens are attracted to root exudates like sugar and amino acids such as the zoospores of many plant pathogens.

ii. Pre-penetration:

This phase includes the growth of the pathogen before its actual entry or penetration into the host. This involves the following steps-

a. Germination of propagules

b. Attachment to the host surface

c. Recognition

a. Germination of propagules: Germination is essentially the change from a low metabolic rate to a high metabolic rate and involves a change from near dormancy to intense activity for which an energy source is required. For germination favourable temperature, humidity, nutrient is required. A film of water or dew drops is essential for germination. The moist condition must last long enough for the pathogen to penetrate or else, the germ tubes dessicate and dry. Many spores have a dormancy period during which they do not germinate. Non-parasitic microbes in the leaf surface, called phyllosphere microbes, are antagonistic and inhibits spore germination of important leaf pathogens. Leaf exudates like malic acid inhibit germination of many spores. When a spore germinates it produces a germ tube. This germ tube must grow and move towards a site through which penetration can take place. This is often helped by chemicals or may be by thigmotropic responses.

b.Attachment: Propagules usually have on their surface mucilaginous sheaths consisting of mixture of polysaccharide, glycoproteins etc which when moisten becomes sticky and help the pathogen to adhere to the plant. Germ tube also get adhered to the cuticular surface by mucilagenous materials. The point of attachment of host tissue and germ tube show degradation due to chemical reaction.

c. Recognition: It is not clearhow the pathogen and host recognise one another. It is assumed that when they come in contact, an early event takes place which triggers a response in either result in disease development or its prevention. This recognition may be self or non-self.

Self-recognition- The pathogen ramifies easily and its growth is not inhibited by the host. This is genetically determined and occurs between susceptible host and virulent pathogen.

Non-self-recognition- This occurs between resistant host and avirulent pathogen. The pathogen acts as a foreign body and impedes its further growth and establishment.

iii. Penetration:

Pathogen penetrate plant surface by direct penetration where they exert effort. Entry might be through natural openings like stomata, lenticels, hydathodes etc or through wounds carried by insects or nematods or man. Some enter only through one way, whereas others enter through more than one. Penetration does not always lead to infection. Many organisms penetrate non-susceptible organisms and thus do not cause disease.

1. Direct penetration through intact plant surface-

It is probably the most common type of penetration in fungi. The protoplasm of the germ tube accumulates at the tip to form a bulbous structure called appresorium. From the under surface of the appresorium a fine hypha called the infection peg (penetration peg) arises which passes through the cuticle and usually the epidermal cell wall. Localised softening of the cuticle occurs facilitating penetration. In its absence, penetration of the cuticle is largely by physical forces. Penetration of the cell wall is usually enzymatic in which pectolytic enzymes and cellulose have an important function.

2. Penetration through wounds:

All bacteria, many fungi, all viroid enter through wounds. These wounds may be natural or may be caused by insects, nematodes or by man. Horticultural practices often involve accidental or even deliberate wounding. The fungi or bacteria apparently germinate on the sap of fresh wounds and subsequently invades adjacent plant cells and establishes itself.

3. Penetration through natural openings:

Many enter through stomata while some enter through hydathodes, lenticels etc. Appressorium is formed in the stomata from where substomatal vesicle is formed in the substomatal cavity. Hyphae then grow from the vesicle and invade cells of the host either directly or through haustoria. Few fungi enter through hydathodes. Penetration through lenticels is also a secondary and less efficient pathway.

4. Penetration through root hairs:

Root hairs are particularly vulnerable to pathogens. *Fusarium oxysporum* is a pathogen which invades through root hair contents of the propagules are injected into unicellular root hairs by a fine puncture or hole. This is also an example of mechanical penetration.

iv. Establishment and Colonisation:

Following penetration, there are many wide variations in the subsequent pattern of development and colonisation of host tissue. In certain pathogens such as powdery mildews, the growth of pathogen is ectotropic. Pathogen causing apple scab, produce mycelium which grows only in the region between cuticle and epidermis. Some produce intracellular mycelium whereas some others produce intracellular mycelium. Infected plants gradually show symptoms of the disease. The time interval between the inoculation and appearance of disease symptoms is called incubation period. During infection pathogens use many chemical weapons like enzymes, toxins, hormones, polysaccharides etc. Enzymes change

the structural integrity of the cell wall, increasing the permeability of cell membrane and directly affects cell protoplast. Growth regulators or hormones alter physiological activities of cells whereas polysaccharides accumulate in the vessels clogging them and cause wilting of the plants. Once the pathogen establishes itself, it gradually overtime proceeds through its reproductive cycle forming secondary propagules which get disseminated and cause secondary infection.

PATHOTOXINS

The term toxin can be defined as a substance generally but not invariably of microbial origin which is involved in host pathogenesis. The credit for the discovery of toxin is generally given to Roux and Yersin who first visualized the presence of a toxin in the disease caused by *Corynebacterium diptheriae.* They are normally proteins which are antigenic in nature. They are different from enzymes as they do not attack the structural integrity of the tissue but affect the metabolism in a subtle manner. They act directly on the cell protoplast.

Toxins may act as poisons on host cell membrane or by inactivating or inhibiting enzyme activity and subsequently interrupting the corresponding enzymatic reactions. Certain toxins act as antimetabolites inducing a deficiency for an essential growth factor.

Toxins may be produced by any microbes; if it is of fungal origin, t is called mycotoxins and the disease they cause are called mycotoxicoses. There are different terminologies to describe toxins (according to Wheeler and Luke, 1963).

i. Phytotoxins: Any compound produced by a microorganism which is toxic to plants is a phytotoxins. They are non-specific , in cited by the pathogens, and show no relation between toxin production and pathogenicity. Eg. Lycomarasmine produced by *Fusarium lycopersici*, Alternaric acid produced by *Alternaria solani*.
ii. Vivotoxin: It is defined as a substance produced in the infected host by the pathogen but is not the initial inciting agent of the disease. There are certain prerequisites a toxin can be called vivotoxin. They are
    a. It must be isolated from the disease plant but should not be present in the healthy plants.
    b. It must be characterized chemically.

c. When introduced in a pure form into a healthy host, it must produce the symptoms of the disease or a portion of the symptoms. Eg; Fusaric acid produced by *Fusarium oxysporum*, Piricularin produced by *Pyricularia oryzae.*

iii. Pathotoxin is a general term used for phytotoxin substance produced by live organisms. They play a causal role in disease production and produce symptoms characteristic of the disease in susceptible plants. They may be produced by the pathogen or by host pathogen interaction. Eg: Piricularin produced by *Pyricularia oryzae*.

iv. Endotoxins is the intracellular toxin formed in bacterial cells and liberated only after their death.

Exotoxins is an extracellular toxin which diffuses from live bacterial cells.

Some toxins produced by phytopathogenic microorganisms asct as general protoplasmic poisons and affect many species of plants of many families. They are general or non-host specific toxins. These have shown to produce all or part of the disease syndrome not only on the host plant but also on other species of plants that are not normally attacked by the pathogen in nature. Non-host specific toxins increase the extent of disease caused by a pathogen but are not essential for the pathogen to cause the disease. There are however, some toxins which are toxic to only a few plant species or varieties, and completely harmless to others. They are host-specific toxins-

Non host specific toxins:

i. Wildfire toxins or tabtoxins- this is produced by the bacterium *Pseudomonas syringae* PV *tabaci.*
ii. Phaseolotoxin caused by *Pseudomonas syringae PV phaseolicola*
iii. Fungi toxin (Tentotoxin)
iv. Alternaric acid produced by *Alternaria solani.*
v. Fusaric acid caused by *Fusarium oxysporum.*

Host specific toxins:

i. HV toxin produced by *Helminthosporiium victoriae*.
ii. AK toxin produced by *Alternaria alternata*.

Non host specific toxins:

1. Wildfire toxin or Tabtoxin:

It is produced by the bacterium Pseudomonas syringae PV tabaci, the cause of wildfire disease of tobacco. The toxin in sterile culture filtrate of the bacterium produced identical symptoms on many other plants also such as bean and soyabean. This toxin is also produced by other pathovars of *P. syringae* such as those occurring on oak, maize and coffee. The toxin is responsible for inducing a characteristic necrotic spot on leaves, the spot surrounded by a yellow halo.

Tabtoxin is a dipeptide of aminoacid threonine and so far are known tabtoxinine. Tabtoxin as such is not a toxin. In the cell it becomes hydrolysed to release tabtoxinine (the active part) which inactivates the enzyme glutamine synthetase. It leads to depleted glutamine level and reduced ability of the plant to defend itself. Due to inactivation of glutamine synthetase ammonia accumulates during photorespiration., thus inhibiting photosynthesis and photorespiration and cholorosis and eventually necrosis.

2. Phaseolotoxin:

   This is produced by the bacterium *P. syringae* PV phaseolicola, the cause of halo blight of bean and some other legumes. Phaseolotoxin is an ornithin-alanin-arginine tripeptide carrying a phosphosulfinyl group. Soon after its production, the plant enzymes cleave, the peptide bonds to release alanine, arginine and phosphosulfinyl ornthin (PSNO), the actual functional moiety of the toxin. The PSNO inactivates the enzyme ornithin carbamoxyl transferase (which converts ornithin to citrulline, a precursor of arginine). The toxin thus cause accumulation of ornithin and depleted level of arginine.

3. Tentoxin:

   This is produced by the fungus Alternaria alternata (previously named *A. tenuis*) which causes chlorosis in seedlings of many plants. Tentoxin is the cyclic tetrapeptide that binds to and inactivates a protein (chloroplast-coupling factor) involved in energy transfer in chloroplasts and inhibits photophosphorylation. It interferes with normal choloroplast development and chlorophyll systhesis, thus inducing cholorosis. It also inhibits the activity of polyphenyl oxydases, involved in defense mechanism of plants.

4. Other non host specific toxins- A range of fungi and bacteria are known to produce several toxins. For eg. Alternaric acid produced by *Alternaria* sp.

Host-specific toxins:

A host specific toxin is a substance produced by a pathogen that at physiological concentrations, is toxic only to the host of that pathogen and shows little or no toxicity against non-host plants. Most host specific toxins must be present for the producing pathogen to be able to cause disease. So far, such toxins are shown to be produced only by some fungi belonging to the genera *Helminthosporium, Alternaria, Phyllosticta, Hypoxylon* although some bacterial polysaccharides of *Pseudomonas* and *Xanthomonas* have also been reported to be host specific.

1. Victorin or HV toxin- This is produced by the fungus *Helminthosporium victoriae,* which appeared in 1945 on the introduced and widely spread variety of oat-victoria and its derivatves. The pathogen infects basal parts of the plant and then to the leaves, causing a leaf blight and destroying the entire plant. The toxin produces histochemical and biochemical changes in the plant, which include changes in cell wall structure, loss of electrolytes from the cell, increased respiration rate, decreased growth and protein synthesis.
2. T-toxin (*Helminthosporium maydis* race T-toxin)- This is produced by the race T of *Helminthosporium maydis*, cause of southern corn leaf blight. The toxin is a mixture of linear, long (35-45 carbon) polyketols. It acts specifically on mitochondria causing early loss of matrix density, rendering them non-functional.
3. AK-toxin- It is produced by a distinct pathotype of Alternaria alternata previously referred to as *A. kikuchiana*, the cause of black leaf spot of Japanese Pear (*Pirus serotina*). The toxin causes the cells to instantaneous lose K-ion and phosphate.
4. AM-toxin- It is produced by the apple pathotype of *A. alternata*, previously known as *A. mali* causing Alternaria blotch of apple. The site and mechanism of action are similar to AK-toxin but this also causes rapid loss of chlorophyll.
5. Other host-specific toxins- Some other host-specific toxin produced by fungi are

- **HC-toxin** produced by *Helminthosporium carbonum* race 1 and the host is corn,
- *H. sacchari* produces **HS toxin** and the host plant being sugarcane,
- *Alternaria alternata* race (f) *lycopersici* produces **AL toxin** and the host plant is tomato.

Phytoalexins:

Phytoalexins are "antibiotics" which are produced as a result of interaction of two metabolic systems, host and parasite and inhibit the growth of microorganism pathogenic to plants.

Phytoalexins are fungistatic rather fungicidal and have no role in viruses, bacteria and nematodes initiated diseases.

Properties of Phytoalexins-

i. The principles designated as phytoalexins inhibit the development of the fungus and activated only when host cells comes n contact with the pathogen.
ii. The defense reactions occurs only in the living cells.
iii. Phytoalexins are inhibitory substances and are the products of secondary metabolism.
iv. Phytoalexins are non-specific in its toxicity towards fungi, however fungal species may be differentially sensitive to it.
v. The basic response that occur in resistant and susceptible varieties is similar and difference lies only in the speed/rate of formation of phytoalexins.
vi. The defense reactions confined to the fungal invaded tissue and its immediate neighbourhood.
vii. The resistant state is not inherited.

Two theories have been suggested to explain the mode of action of phytoalexins-

1. Differential sensitivity theory-
   Both pathogenic and non pathogenic fungi as well as saprophyte can induce the production of phytoalexin in both resistant and less sensitive varieties. But pathogenic races are less sensitive to the toxicity of phytoalexins produced by the susceptible host and can cause disease.

Eg. Pea produces pisatin. *Ascochyta pisi* can tolerate pisatin and cause disease in susceptible varieties. *Fusarium solani* is non pathogenic and is destroyed by the pisatin even in susceptible variety.

2. Differential synthesis theory –
   Pathogenic fungal sp induces production of phytoalexins in low concentration than non pathogens.
   Eg. Different varieties of pea produce different concentration of pisatin when inoculated with *Ascophyta pisi*.

Examples-

Phytoalexin	**Host**	**Pathogen**
Glycollin	Soybean (*Glycine max*)	*Phytopthora megasperma* var. *sajae*
Phaseollin	Bean (*Phaseolus vulguris*)	*Monilina fructicola*
Medicarpin	*Vicia faba*	Botrytis cinerea
	Medicago sativa	Stemphylium botysum
	Trifolium pratense	Helminthosporium furcicum
Pisatin	*Pisum sativum* (Pea)	*Ascochyta pisi*
		Monilina fructicola
Rishitin	*Solanum tuberosum*	*Phytopthora infestans*
Gossypol	Cotton (*Gossypium* sp)	*Verticillium alboaratrum*
		Rhizopus migricans
Momilactone A or B	Rice (*Oryza sativa*)	*Pyricularia oryzae*