

## **Lecture 1**

### **Plant disease epidemiology – Meaning and importance, difference between simple and compound interest diseases – Factors affecting plant disease epidemics – host, pathogen, environment and time factor**

Epidemiology or epiphytology is the study of the outbreak of disease, its course, intensity, cause and effects and the various factors governing it. Based on the occurrence and geographical distribution they are classified as follows:

#### **Endemic or Epiphytotic**

When a disease is more or less constantly occurring year after year in a moderate to severe form in a country or locality then it is called an endemic disease. eg: wart disease of potato (*Synchytrium endobioticum*) is endemic in Darjeeling, citrus canker (*Xanthomonas axonopodis* pv *citri*) in Asia and sorghum rust (*Puccinia purpurea*).

#### **Epidemic or Epiphytotic**

It is a sudden outbreak of a disease periodically over a widespread area in a devastatingly severe form causing severe losses or complete destruction. This is constantly present in a locality but it assumes severe form only on occasions. This is because of the occurrence of favorable environment responsible for the rapid development of disease. eg: wheat stem rust (*Puccinia graminis tritici*) and powdery mildew (*Erysiphe graminis v. tritici*), late blight of potato (*Phytophthora infestans*), red rot of sugar cane (*Colletotrichum falcatum*), downy mildew of grapevine (*Plasmophora viticola*) and rice blast (*Pyricularia oryzae*).

Certain diseases are endemic in one area and become epidemic in another area. Eg: Citrus canker is endemic in Asia but epidemic in the introduced place, Florida (U.S.A). The downy mildew of corn is an endemic disease in India but became epidemic in the Philippines.

#### **Pandemic**

When an epidemic disease spreads over continents or subcontinents and involves mass mortality it is considered as pandemic. The outbreak of black stem rust of wheat in India during 1947 is the best example for a pandemic disease.

#### **Sporadic**

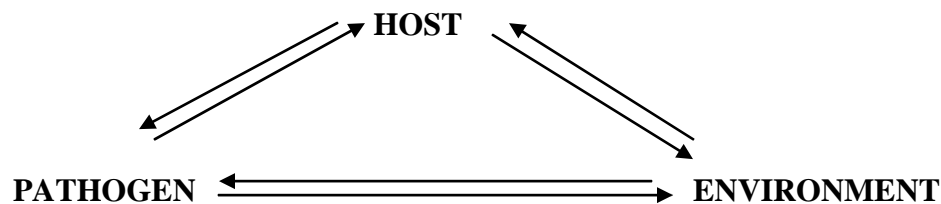
Diseases which occur at irregular intervals over limited areas or locations are called sporadic. They occur relatively in few instances. Eg: Fusarium wilt of cotton (*Fusarium*

oxysporum f. sp. vasii infectum) grains mut of sorghum (*Sporisorium sorghi*) and loose smut of wheat (*Ustilago nuda*).

An epidemic may cause widespread and mass destruction of crop in a short time or may persist for long periods depending upon the three following factors responsible for the disease:

1. Host
2. Pathogen and
3. Environment

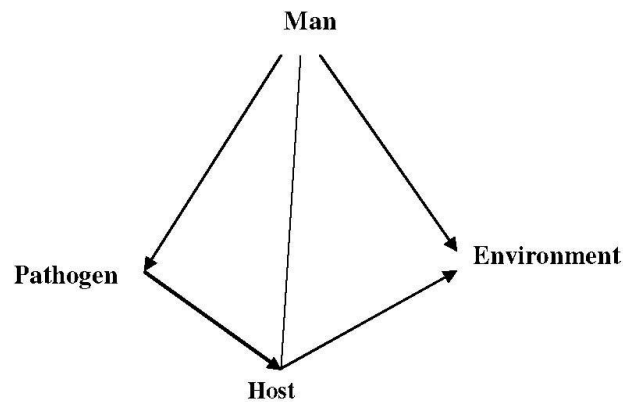
**Environment flowchart**



**Pathogen**

A course of epidemic in nature differs with the nature of the host, the pathogen and the environment. In arecanut the Koleroga fungus, *Phytophthora arecae* become destructive during monsoon period (July-Sep) and wanes away with rising temperatures and dry conditions. The above disease once again become destructive during rainy season. This type of epidemic is known as seasonal epidemic or annual epidemic. Outbreak of *Phytophthora* wilt of betel vine occurs during rainy season in South India. In temperate zone peach leaf curl and apple scab follow the similar course.

Epidemics caused as a result of introduction of new pathogens in the locality hitherto free from them, appear in two phases viz., destructive phase and innocent phase (due to biological equilibrium reached between new comer pathogen and the original inhabitant). The well known epidemics of late blight of potato in Europe and blast disease of rice in South East Asia, powdery mildew and downy mildew of grapevine in Europe, leaf rust of coffee in Sri Lanka and anthracnose of grapevine in India are examples of this category. In the above diseases the pathogens after taking heavy toll of the crops have settled down.



**Factors governing epidemic or essential conditions for an epidemic**

A disease is sometimes sporadic and assumes epidemic proportions under special circumstances. The essential conditions for an epiphytotic or the factors governing epidemics can be grouped under the three heads.

1. Nature of host
2. Nature of the pathogen and
3. Environment

An epidemic can only result from the cumulative effects of all the three factors mentioned above, acting simultaneously. Few pathogens are capable of assuming epiphytotic conditions while others are sporadic. The former group consists of late blight of potato, blast of rice, downy mildew diseases and rust diseases.

<b>Host</b>	<b>Pathogen</b>	<b>Environment</b>
Susceptibility of the host	Introduction of a new Pathogen	Temperature
Aggregation and distribution of susceptible hosts	Presence of aggressive train of the pathogen	Moisture and humidity
Introduction of new hosts	High birth rate of the Pathogen	Rainfall
Introduction of	Low death rate of the pathogen	Light and

new collateral or alternate pathogen host		shade
	Easy and rapid dispersal of the pathogen Adaptability of the pathogen	Wind

**A. Host Factors**

**1. Susceptibility of the host**

Plants have ability to combat disease which manifests itself as susceptibility or resistance. Plants are predisposed to the attack depending on their nature, environment and stage of growth. Presence of susceptible varieties in an area may act as one of the causes of epidemic. For example, late maturing varieties of groundnut are more susceptible to early leaf spot (*Cercospora arachidicola*) and late leaf spot (*Phaeoisariopsis*) than the early maturing varieties. Similarly late maturing varieties of wheat are susceptible to loose smut (*Ustilago nuda tritici*) than the early maturing varieties. Early sown sugarcane varieties of sugarcane are more susceptible to leaf rust in Deccan canals in Bombay area than the late sown varieties.

Wheat plant becomes susceptible to black rust (*Puccinia graminis tritici*) at the boot stage but is resistant when young. Susceptibility of rice plants to blast disease (*Pyricularia oryzae*) increases with application of heavy doses of nitrogenous fertilizers. Cottons plants are susceptible to *Fusarium* wilt (*F. oxysporum f.sp. vasinfectum*) at soil temperatures of 26 to 28°C, brinjal to *Verticillium* wilt *Verticillium dahliae* at 20°C. But crop plants are resistant to these soil-borne diseases at relatively lower or higher temperatures. Under the above conditions, the pathogen multiplies faster, cause infection and effectively uses its propagules for quick secondary spread causing epidemic.

**2. Aggregation and distribution of susceptible hosts**

Abundance of susceptible hosts in an area is one of the major causes of the spread of epidemics. Continuous cultivation of susceptible variety or varieties in an area, that too in a large contiguous area help in the build up of inoculum and improve the chances of epidemics. Under the above conditions the pathogen increases the rate of multiplication of its propagules and repeats the disease cycles in a short span. Wheat cultivation area in the U.S.A and Canada and

rice cultivation area in East Asian countries are exposed to a greater danger of epidemics by wheat black rust and rice blast respectively.

Destructive epidemic of early and late leaf spots of groundnut in Bombay area (Gujarat and Maharashtra States) during 1912-1913 was mainly the result of cultivation of local varieties in a larger area. Panama wilt (*Fusarium oxysporum* f.sp. *vasinfectum*) susceptible table variety, 'Son' in banana was responsible for the destructive epidemic in parts of Bombay area (Gujarat and Maharashtra) during 1936-

1940 Countrywide cultivation of red rot (*Colletotrichum falcatum*) susceptible sugarcane varieties (local varieties like *Pundya*, *Khajuria* etc.) practically made their cultivation impossible in Bombay area.

### **3. Introduction of new host(s)**

Disease proneness in the host is induced by environment and other factors. The host is liable to vigorous attack and successful infection by the pathogen. A resistant or moderately resistant variety may become susceptible or highly susceptible. A susceptible variety may become highly susceptible when conditions favouring proneness are existing and cause severe damage. Under the above conditions the pathogen multiplies faster, cause infection and produces more propagules for secondary spread. Introduction of an exotic cotton variety (C4 (Cambodia)) caused outbreak of bacterial blight (*Xanthomonas axonopodis* sp. *malvacearum*) and grey mildew (*Septoacylindrium gossypii*) in local variety, Deviraj, grown in Maharashtra area in India.

### **4. Introduction of new collateral or alternate hosts**

Alternate hosts are those plants on which the heteroecious pathogens pass part of their life cycles. Similarly, collateral hosts are some wild plants in which the pathogen survives when primary host is not available. Both alternate and collateral hosts are important in building up the primary inoculum to the next crop. They determine the course and intensity of an epidemic.

Grass hosts (collateral hosts) of *Sclerosporasacchari*, *S.philippinensis* (downy mildews), *Pyricularia oryzae* (rice blast), *Ustilago scitaminea* (sugarcane smut) may produce abundant inoculum which aid in building up of epidemics. Outbreak of heteroecious blister rust of pine (*Cronartium ribicola*) in Europe and the U.S. happened due to import or introduction of *Pinus strobus* from the USA.

## **B. Pathogenic Factors**

### **1. Introduction of new pathogen**

Some pathogens, epidemic in certain area, may become quite aggressive and outbreak as epidemic when introduced to new area. For example late blight of potato caused by *Phytophthora infestans* was epidemic in South America. This disease became epidemic when the infected tubers were introduced in Europe (in 1843-45). Fire blight (*Erwinia amylovora*) in North America is endemic. Fire blight spread to Pacific coast fruit-growing areas of the U.S.A in 1884 and subsequently it reached Canada. It reached New Zealand in 1919 and it appeared in England in 1957. The mode of introduction had been through fruit boxes. Coffee rust (*Hemileia vastatrix*) is indigenous in Ethiopia, where *Coffea arabica* is native. The disease spread to Sri Lanka in 1869, India in 1870, Sumatra in 1876, Java in 1878 and the Philippines in 1889. It also spread from Kenya to the Congo by 1918 and reached the Cameroons. From 1950 onwards, it spread to the remainder of West Africa.

The mode of long distance transport of *H. vastatrix* is wind. Spores have been trapped at up to 1000 m above sea level up to 150 m from infected sites. Dutch elm diseases (*Ceratocystis ulmi*) first reported in 1919 in Holland, spread throughout Europe and reached Great Britain in 1927. It was introduced to the eastern United States on elm logs imported from Europe.

### **2. Presence of aggressive strain of the pathogen**

All the strains of a pathogen are not aggressive. Only the aggressive strains are capable of causing infectious diseases which spread as epidemic. They are characterised by rapid cycle of infection and causing successful infection in new hosts. Rapid cycle of infection is essential for successful infection and it happens only by aggressive strain of the pathogen. e.g., *Puccinia graminis tritici* (wheat black rust) in India, stripe rust, bunt and loose smut of wheat in the U.S.A. and Europe. The possibility of outbreak of epidemics increases with the number of physiologic forms or pathogenic strains of the pathogen present in a locality.

### **3. High birth rate of the pathogen**

Pathogen with high reproductive capacity and capable of rapid dissemination over wide areas mostly cause epidemics. The fungal members causing powdery mildews, downy mildews, rusts, blights, etc., produce enormous amount of spores. These spores are easily dispersed by using water or insects and cause infection to new plants. The high degree of fecundity and

the enormous amount of inoculum produced by some common plant pathogens are given in table.

**Fecundity rates of plant pathogens**

Sl. No	Pathogen	Extent of fecundity
	Wheat stem rust ( <i>Puccinia graminis tritici</i> )	Twenty five trillion uredospores in one hectare of wheat crops.
2	Wheat stem rust ( <i>Puccinia graminis tritici</i> )	64,000 million aeciospores from aecial cups in a single barberry bush
3	Cedar rust of apple	Two billion teliospores in a single gall.
4.	Acorn plant infected with downy mildew	225 million sporangia in one night
5.	A. grapevine infected by downy mildew	32,000 sporangia per sq. cm
6.	Bunt of wheat	6 to 12 million smut spores in a single kernel
7.	Smut of corn	125,000 billion smut spores in one hectare
8.	Chestnut blight	150,000,000 spores in a single spore horn
9.	<i>Fomes applanatus</i>	5,460 billion spores in a single fruiting body

**4. Low death rate**

Epiphytotic may also be caused by low death rate diseases. These diseases are caused by agents of systemic nature which are protected by plant tissues. As they are protected by plant tissues the chances of high mortality is reduced to the minimum. In these diseases the chief source for accumulation of inoculum for epiphytotic is the diseased plant organ used for vegetative propagation (corms, setts, tubers, etc.). Here the build up of epidemics is comparatively low compared to high birth rate diseases. When a particular area is planted and covered with diseased planting material the chances of occurrence of epiphytotic are very high. e.g., virus and phytoplasma diseases in crops propagated through vegetative plant parts.

### **5. Easy and rapid dispersal of the pathogen**

The ability of the pathogen to cause epidemic depends both on the high birth rate and dispersal. The propagules of the pathogen produced should be dispersed for development of an epidemic. It may happen by external agencies like wind, water, insects, mites and nematodes. Fungal spores / conidia are minute and light and resistant to adverse conditions. Fungal spores are mostly disseminated by wind. Bacteria are mostly disseminated by water or insects. Virus and phytoplasma diseases are mostly transmitted by insects, mites or nematodes. Epidemics are determined by the velocity of wind, direction of wind, moisture, relative humidity, temperature, presence and number of vectors and their rate of reproduction.

### **6. Adaptability of the pathogen**

Pathogens have the capacity to adapt to adverse conditions. Fungi produce different types of spores like oospores, ascospores and smut spores (chlamydospores) which help in tiding over adverse conditions. Bacteria survive in diseased plant parts. Viruses and phytoplasmas live in collateral hosts or insect vectors in the absence of the suitable crop hosts.

### **C. Environmental Factors**

The environmental conditions such as temperature, relative humidity, rainfall, duration and intensity of light, etc. play very important role in causing epidemics. These are actually the deciding factors and influence almost all the stages of disease cycle. Favourable environmental conditions are needed for sporulation, liberation of spores, dissemination of pathogen, germination, infection and establishment of pathogen in the host.

For example, persistent optimum temperature and moisture are needed for spore germination and entry of germ tube in the host. Similarly optimum temperature, moisture, light and specific nutrition is required for the development of the disease and sporulation of pathogen. Compound interest diseases and simple interest diseases. The terms compound interest and simple interest are for explaining rate of increase of pathogen. These terms were introduced by Vander Plank in 1963 in the book 'Plant Diseases -

Epidemics and Control'. Based on the mode of multiplication of pathogen, the diseases are classified into two types:

1. Simple interest diseases
2. Compound interest diseases



### **1. Simple interest diseases**

In simple interest diseases the increase is mathematically analogous to simple interest in money. There is only one generation of the pathogen in the life of the crop. The primary inoculum is seed-borne or soil-borne. The secondary infection rarely occurs during the crop season. That is, the pathogens do not spread from plant to plant in one growing season. Simple interest diseases are caused by seed- or soil-borne smuts, like loose smut of wheat, covered smut of barley and soil-borne fungi which attack roots, like wilt (*Fusarium oxysporum*) and root rot (*Rhizoctonia* spp.) diseases.

Most of the smuts infect the seedlings, grow along with the growth of the plant and produce spores in the inflorescence on maturity of the crop. There is no secondary spread from the smutted heads. These smut diseases are mostly systemic in nature. They do not produce propagules external to the host during the active season of the crop. Dispersal of propagules of these fungi is restricted by existing climatic and biotic conditions.

### **2. Compound interest diseases**

In compound interest diseases the rate of increase is mathematically analogous to compound interest in money. The pathogen produces enormous amount of spores at a very rapid rate. These spores are disseminated rapidly by wind and infect the other plants. Both the inoculation and sporulation period are short so that the pathogen spreads from plant to plant during the same growing season. New crop of spores is produced, disseminated and the cycle is repeated fast. Thus more generations of the pathogen are produced in the life of a crop. e.g., late blight of potato, powdery mildews and rust diseases. If we consider wheat stem rust caused by *Puccinia graminis tritici* as an example, the fungus produces uredospores in very large numbers (50,000 to 4,00,000 uredospores per uredosorus).

These spores are spread by wind and infect other plants. Each of the freshly infected wheat plant produces uredopustules within 5 to 7 days at 24°C. Thus within a week of appearance of the first pustule in the crop several thousand new pustules are formed which could repeat the process within a week. If the climatic conditions of about 24°C temperature and relative humidity remain for only few weeks, the entire crop is severely affected by the disease.

### **Course of epidemic**

The course of epidemic follows two distinct phases viz.,

- i. Progressively destructive phase and

ii. the decline phase

**i. Progressively destructive phase**

Some epidemics develop slowly (tardive) while others develop rapidly. Slow epidemics (or epiphytotics) usually occur among population caused by systemic pathogens. The pathogen multiplies slowly following the characters of simple interest disease. They belong to low

death rate category and have less incubation period and sporulation period. However, their epiphytotic nature is greatly influenced by environmental factors.

**ii. Decline phase**

During early stage, an epidemic spreads vigorously causing diseases in new hosts. After development of a saturation stage it shows a decline by itself. No epidemics may be due to non-availability of susceptible stages of the crop, unfavourable weather conditions and reduction in aggressiveness of the pathogens. Generally the hosts are prone to the disease at a specific developing stage. Once this stage is crossed in a plant its proneness to infections is reduced or completely lost. Under the conditions the epidemic declines. The decline in the epidemic may also be due to unfavourable weather conditions for disease development. As a result future spread of the disease will be checked and the epidemic will decline. Wheat crop in Northern India usually gets the attack of rusts in January to March.

Epidemics develop during these months. Although the plant remains prone to attack afterwards also, further development of the disease is checked because of rise in temperature which is favourable for the pathogen. Due to the above mentioned and other causes, the aggressiveness of the pathogen may be reduced. When all susceptible individuals are destroyed by the pathogen, it may try to parasitize the remaining resistant individuals of the same species. In these adverse conditions, the pathogen may lose its power of successful infection, its reproduction may slow down and the pathogen becomes less aggressive.

**Slow and rapid epiphytotics**

The form of epidemic is decided by the nature of the pathogen, host and the weather. Epidemic may develop slowly and is called 'tardive'. Epidemic which develops rapidly is called 'explosive'. In between these intermediate forms of epidemic may occur.

**i. Slow epiphytotics**

Slow epiphytotics occur among perennial (tree) populations. Infected host survives for several years before dying. Most of the characters of a simple interest disease are found in slow

epiphytotic. The causal agent is mostly systemic. The pathogen multiplies slowly. Their movement from plant to plant is much slower. They are low death rate pathogens. In slow epiphytotic, crop sanitation is the best method. e.g., Swollen shoot of cocoa.

This disease spreads very slowly from tree to tree and still less from one garden to another garden. For instance, the incidence of 31 % swollen shoot increased to 75 % over a period of 2.5 years. As stated by Van der Plank (1959) the rate of multiplication of a systemic disease of trees is about ten fold a year whereas it is 10,000 fold in respect of herbaceous plants and it is of higher rates for local lesion pathogens e.g., late blight of potato, wheat stem rust, etc.,

## **ii. Rapid epiphytotic**

Rapid epiphytotic occur among annual crops. It is caused by non-systemic pathogens with high birth rate. Several generations of the pathogen is produced within a short time.

Rapid epiphytotic are largely governed by environmental factors compared to slow epiphytotic. Disease increase is rapid and the disease rises to a peak in short time and then show sharp decline when the weather turns unfavourable or when the host becomes resistant due to maturity or due to restricted dispersal of propagules of pathogen. e.g., apple scab. This type of epiphytotic is controlled by protective spraying or dusting with chemicals.

**Plant Disease Forecasting – Meaning, advantages, methods in forecasting and examples****Disease Forecasting**

Forecasting of plant diseases means predicting for the occurrence of plant disease in a specified area ahead of time, so that suitable control measures can be undertaken in advance to avoid losses. Disease forecasts are predictions of probable outbreaks or increase in intensity of disease. It involves well organized team work and expenditure of time, energy and money. It is used as an aid to the timely application of chemicals. Among the first spray warning services to be established for growers, were the grapevine downy mildew forecasting schemes in France, Germany and Italy in the 1920s. Disease forecasting methods are available for the following plant diseases.

Sl. No	Plant disease	Countries
1.	Grapevine downy mildew	Australia, France, Germany, Greece, Italy, Romania, Spain, USSR, Yugoslavia
2.	Cucurbit downy mildew	U.S.A.
3.	Potato late blight	Australia, Brazil, Finland, France, Germany, Greece, Japan, the Netherlands, Norway, Peru, U.K, the U.S.S.R.
4.	Tobacco blue mould	Canada, U.S.A.
5.	Apple and pear scab	Australia, Canada, Netherlands, New Zealand, U.S.A.
6.	Sugar beet root rot ( <i>Aphanomyces</i> sp.)	U.S.A
7.	Wheat brown (Leaf) rust	U.S.A.
8.	Corn bacterial wilt ( <i>Erwinia stewartii</i> )	U.S.A
9.	Sugar beet curly top	U.S.A.

### **Information needed for disease forecasting**

Forecasting diseases is a part of applied epidemiology. Hence, knowledge of epidemiology (development of disease under the influence of factors associated with the host, pathogen) is necessary for accurate forecasting. The factors of epidemic and its components should be known in advance before forecasting is done.

The information required for forecasting are:

#### **1. Host Factors**

- a. Prevalence of susceptible varieties in the given locality
- b. Response of host at different stages of the growth to the activity of pathogen e.g. Some diseases are found during seedling stages while others attack grown-up plants and
- c. Density and distribution of the host in a given locality. Dense populations of susceptible variety invite quick spread of an epidemic. Growing susceptible varieties in scattered locations and that too in a limited area are less prone to epiphytotic.

#### **2. Pathogen factors**

- a. Amount of primary (initial) inoculum in the air, soil or planting material
- b. Dispersal of inoculum
- c. Spore germination
- d. Infection
- e. Incubation period
- f. Sporulation on the infected host
- g. Re-dispersal/Dissemination of spores
- h. Perennating stages
- i. Inoculum potential and density in the seed, soil and air

#### **3. Environmental factors**

- a. Temperature
- b. Humidity
- c. Light intensity
- d. Wind velocity

### **Requirements or conditions for disease forecasting**

There are five main requirements which must be satisfied before a useful and successful disease forecast is made.

1. The disease must cause economically significant damage in terms of yield loss or quality. Damage assessment is essential to develop strategy for controlling a disease. e.g., Annual estimation of yield loss caused by barley powdery mildew (*Erysiphe polygoni*) in England and Wales had ranged from 6 to 13 %. Potato late blight can cause a yield loss of 28% if the disease reaches the 75% stage by mid- August. Diseases like apple scab and potato common scab reduce the quality of the produce, lower the value of the harvested crop and cause considerable financial loss to the growers.

2. Control measures must be available at an economically acceptable cost.

3. The disease must vary each season in the timing of the first infections and its subsequent rate of progress. If it does not, there is no need for forecasting.

4. The criteria or model used in making a prediction must be based on sound investigational work carried out in the laboratory and in the field and tested over a number of years to establish its accuracy and applicability in all the locations where its use is envisaged.

5. Growers must have sufficient man power and equipments to apply control measures when disease warning is given. Long-term warnings or predictions are more useful than short-term warnings or predictions.

### **Methods of disease forecasting**

Disease forecasting requires field observations on the pathogen characters, collection of weather data, variety of the crop and certain investigations and their correlations. Usually the following methods are employed in disease forecasting.

#### **1. Forecasting based on primary inoculums**

Presence of primary inoculum, its density and viability are determined in the air, soil or planting material. Occurrence of viable spores or propagules in the air can be assessed by using different air trapping devices (spore traps). In the case of soil-borne diseases the primary inoculum in the soil can be determined by monoculture method.

Presence of floes of smut of wheat, ergot of pearl millet and viral diseases of potato can be detected in theseed lots at random by different seed testing methods. Seed testing methods can be used to determine potential disease incidence and enable decision to be made on the need for chemical seed treatment. The extent of many virus diseases is dependent on the severity of the preceding winter which affects the size of vector population in the growing season. e.g., Sugarbeet yellow virus.

## **2. Forecasting based on weather conditions**

Weather conditions viz., temperature, relative humidity, rainfall, light, wind velocity etc., during the crop season and during the inter crop season are measured. Weather conditions above the crop and at the soil surface are also recorded.

## **3. Forecasting based on correlative information**

Weather data of several years are collected and correlated with the intensity of the diseases. The data are compared and then the forecasting of the disease is done. Forecasting criteria developed from comparisons of disease observation with standard meteorological data have been provided for diseases like *Septoria* leaf blotch of wheat, fire blight of apple and barley powdery mildew.

## **4. Use of computer for disease forecasting**

In some advanced countries forecasting of disease is made by the use of computers. This system gives the results quickly. One such computer based programme in the USA is known as 'Blitecast' for potato late blight. Examples of well developed forecasting systems are given below.

### **a. Early and late leaf spots of groundnut**

A technique has been developed for forecasting early and late leaf spots of groundnut in the U.S.A. When the groundnut foliage remains wet for a period greater than or equal to 10 h and the minimum temperature is 21°C or higher for two consecutive days or nights, the disease development is forecasted.

A computer programme has been developed in the USA. This is accurate and is widely used in the USA. The data on hours for day with relative humidity (RH) of 95% and above and minimum temperature (T) during the RH observations for the period, for the previous 5 days are fed to the computer. Calculations are rounded to whole numbers. The T/RH index for each of the five days is calculated e.g., when hours of the RH 95% equal 10 and the minimum temperature during the period equals 21.1°C the T/RH index is 2.0. The T/RH indices for days 4 and 5 are summed. If the total index exceeds 4 disease is forecasted. If the index is 3 or less no disease is forecasted.

### **b. Late blight of potato**

In the USA a forecasting programme has been developed for late blight of potato (*Phytophthora infestans*). The initial appearance of late blight is forecasted 7 to 14 days after the occurrence of 10 consecutive blight favourable days. A day is considered to be blight favourable when the 5 day average temperature is 25.5°C and the total rainfall for the last 10 days is more than 3.0 cm. A computerized version (Blitecast) has also been developed in the U.S.A for forecasting potato late blight. Blitecast is written in Fortran IV. When a farmer desires blight cast (blitecast) he telephones the blight cast operator and reports the most recently recorded environmental data. The operator calls for the blight cast programmes in the computer viz., typewriter terminal and feeds the new data into the computer. Within a fraction of second the computer analyses the data and series of a forecast and spray recommendations to the operator who relays it to the farmer.

The entire operation can be completed during standard three minutes telephone call. The system makes one of the four recommendations viz., no spray, late blight warning, 7 days spray schedule or 5 days spray schedule. The last 5 days spray schedule is issued only during severe blight weather. In West Germany, 'Phytoprog' is the programme used. It is based on measurements of temperature, relative humidity and rainfall. Phytoprog provides a negative prognosis (an indication of when the usual routine spray application should be dispensed with).

### **c. Blister blight of tea**

A system for predicting epidemics of blister blight of tea (*Exobasidium vexans*) has been developed based on the number of spores in the air in the tea plantation and the duration of surface wetness on the leaves. The duration of sunshine is negatively correlated with the duration of surface wetness. The following prediction equation has been developed.  $Y = 1.8324 + 0.8439X_1 + 0.9665X_2 - 0.1031X_3$  where,  $X_1 = \log \% \text{ infection } t_2$ ,  $X_2 = \log \% \text{ infection } t_1 - \log \% \text{ infection } t_2$ ,  $X_3 = \text{mean daily sunshine for a 7 days period preceding } t_2$

### **d. Southern corn leaf blight**

'Epimay' is a system for forecasting Southern corn leaf blight (*Bipolaris maydis*) based on conceptual model.



**e. Riceblast**

In India, forecasting rice blast (*Pyricularia oryzae*) is done by correlative information method. It is predicted on the basis of minimum night temperature 20 to 26°C in association with high relative humidity of 90% or above. Computer based forecasting system has also been developed for rice blast in India.

**f. Wheat stem rust**

Forecasting wheat stem rust epidemic is done by analysing the rain samples which give precise data for inoculum present in the air. Moreover several wind trajectories are also prepared to survey the air-borne primary inoculums and its deposition. It has been observed that primary inoculum comes from South India, to the plains of Central and North India.

**g. Brown striped downy mildew of corn**

The forecasting of brown stripe downy mildew of corn (*Sclerophthora rayssiae* var. *zeae*) which is restricted to India is done on the basis of average rainfall 100 to 200 cm or more accompanied by low temperature ( 25°C or less). Spore trapping Techniques of acquisition of biological data for consecutive forecasting models are important. Spore traps have been widely used into completed disease with weather conditions.

Spore trapping is useful for understanding epidemiology of a disease and behaviour of the pathogens. This helps in developing models on dispersal of pathogens or on epidemiology of the disease and to formulate methods of management. Methodology of spore trapping depends on the following objectives of the worker.

1. Biology of the pathogen
2. For infection forecasting
3. Spore dispersal gradients
4. Management of the disease

In epidemics of air-borne plant diseases the number of spores of the pathogen landing on the plant which depends on the number of spores in the atmosphere above the crop is an important factor for the quantitative sampling of the atmosphere (number of spores per unit volume of air). For trapping and estimating these studies different types of traps are used. The following spore traps are usually employed in trapping of fungal spores.

Cylindrical rods or microscopic glass slides: It helps to gather data on the spore arrival in a locality. In this, the surface of microscopic slide is smeared with grease and made sticky. In this method, quantitative estimation is not possible as the number of spores collected is very low.

### **1. Hirst's volumetric spore trap (Hirst 1952)**

In this instrument, air is sucked into at a controlled rate and impinged on to a glass slide moved by a clockwork mechanism past the orifice. It gives continuous count of spores in 24 h. The number of spores per unit volume of air at any given time can thus be calculated.

### **2. Rotorod sampler or rotorod spore trap (Sutton and Jones 1976)**

It comprises of a 'U' shaped rod attached at its mid point to the shaft of a small battery operated electric motor. In this equipment the surface of the rod is covered with a vaseline strip of transparent cellophane to catch spores which can be taken off and mounted on a glass slide. From the area of the strip and the speed of rotation, the volume of air samples can be calculated.

### **3. Anderson cascade spore sampler**

It is a device where Petri plates with nutrient agar are used to collect the spores.

### **4. Bourdillon slit sampler**

Air is sucked in a chamber by vacuum pump which strikes the rotating Petri dish containing agar medium. The agar medium retains the spores sucked in the air. Concentration of viable spores is calculated after counting germinated spores in the medium.

### **5. Burkard's 7 day volumetric spore trap**

This device records spores in the air drawn by a pump on 7 days basis on a cellophane strip wrapped on a drum rotating inside a chamber.

### **6. Jet spore trap**

In the above sampling methods, the viability of the spores cannot be determined. To overcome this, living plants have been used as spore traps. A jet spore trap in which spores are impacted in an air jet into a column of still air, through which they fall, to settle on leaf segments exposed at the base of the chamber. In this trap, suitable cultivars of host plants can be employed to determine number of viable spores.

**Remotesensing–Meaning,scope,objectives,advantages**

Remote sensing carries many different connotations to different individuals, ranging from photography to large satellite platforms. Each day we are provided many frames of remotesensing information through our eyes, which we use to make visual assessments of an object. These scenes provide an information source about objects from which we judge certain characteristics, e.g., size, condition, or change. The local TV weather report uses remote sensing of clouds to show the passage of storms. Plant pathologists have used remote sensing tools for a number of years and were among the first to use color infrared photography to assess the presence of disease in trees. The application of remote sensing via airborne cameras provided an answer to a question that would not have been possible through ground surveys. In many aspects we have progressed rapidly to our current state of knowledge about the utility of remote sensing. The intent of this address is to arouse the interest of individuals in discovering how remotesensing could be applied to plant pathological problems of today and tomorrow.

**Regions of The Spectrum**

The spectrum of electromagnetic radiation ranges from the short, high energy wavelengths to the long radio waves. As a receptor, the human eye only measures a relatively small portion of the spectrum in the visible wavelengths from 0.4 to 0.7  $\mu\text{m}$ . Remote sensing instruments, on the other hand, have utilized wavelengths extending in the microwave region for a variety of applications. For this discussion, we will confine the wavelengths to the region from 0.4 to 14  $\mu\text{m}$ . The region from 0.4 to 5  $\mu\text{m}$  can be represented as the reflected wavelengths. Reflection is that phenomenon in which an impinging beam of radiation of a particular wavelength is reflected back away from the object without any change. This can be contrasted to emittance, which is the emitting of radiant energy at a particular wavelength due to the temperature of an object. Surfaces at the temperature of the earth (300°K) emit mostly in 10-12  $\mu\text{m}$  waveband, while the sun at 6000°K emits in the 0.5  $\mu\text{m}$  region. Both reflectance and emittance provide information that can be utilized in applying remote sensing to agricultural problems.

**Reflection from Leaves**

Reflection from individual leaves is not constant across the wavelengths from 0.4 to 2.5  $\mu\text{m}$ . Leaves have a low reflectance in the visible (0.4-0.7  $\mu\text{m}$ ), a high reflectance in the near-

infrared (0.7-1.2 Mm), and a low reflectance in the middle and far infrared (1.2 Mm) wavebands. This variation in leaf reflectance has allowed for the differentiation of leaves from soil, which tends to show little variation in reflectance across these wavelengths.

Reflectance from leaves is species dependent and sometimes cultivar dependent. The primary variation among species is in the visible reflectance and is due to species or leaf age. Reflectance tends to increase in individual leaves as the leaf matures; however, the changes are wavelength dependent. These changes are due to changes in intracellular water content and chlorophyll content. Lesions and reduction in chlorophyll content created by a disease also cause an increase in reflectance. Water stress by reducing the internal water content increases the reflectance from an individual leaf. Information gathered from individual leaves provides a basic set of information about the mechanism of the changes occurring within a plant; however, to be of practical application it must be extended to a canopy or field level.

### **Reflection from Canopies and Fields**

Composites of leaves or canopies exhibit the same reflective properties of individual leaves; however, there are a series of variables that now must be considered. Leaf orientation, i.e., the arrangement of leaves on the stem and orientation to the sun, provides a source of variation when viewing a canopy compared with an individual leaf. Also, all leaves are not exposed to the same level of incoming radiant energy and often do not reflect back to the sky due to distortions in the leaf surface. Leaf surfaces often act as polarizing filters and reflect back to portions of the sky that are not always detected by viewing the canopy only the vertical direction. However, the information contained in bidirectional and polarized reflectance has yet to be fully exploited in the evaluation of canopy response to stress. Leaf fluorescence is another attribute that has been observed in all plants and can be related to the efficiency of the photosynthetic process. It is possible that leaf fluorescence could be used to assess the impact of diseases on the physiological status of the plant. This technique has only been used on individual leaves; however, it could be extended to canopies through the use of laser-induced fluorescence. This procedure will have to be adapted to plant canopies but may become a powerful and useful research tool.

Canopies of plants are grown in fields with varying soil, and soil also has some unique reflective properties. The variation across wavelengths is less for soil than for leaves; however, the reflectance changes in response to modifications in the surface. The addition of organic

matter as residue on the surface reduces the reflectance. Soils vary in reflectance due to mineral composition and weathering of the minerals. However, across the visible and near-infrared wavelengths the reflectance from soil remains relatively constant within a given soil type. Change in water content in the upper 2 mm causes the largest variation in reflectance. Water has a low reflectance and the addition of a water film around the soil aggregate causes an increased absorption of the incident radiation. As a soil is wetted there is a darkening in the color, which lightens as the soil dries. This variation in the reflectance from soils due to changing soil water adds, complexity to the reflectance from canopies, particularly when there is less than complete ground cover by the plant, i.e., exposed soils when viewed from above the plant. Since there is a changing amount of plant material both in the adding of new leaves or the senescence of the older leaves, there is a continually changing scene to be viewed. This challenge must be faced and understood if we are to develop the tools that allow us to assess the effect of a disease or any other stress on the plant.

Instruments available for the measurement of reflected radiation adaptable to remote sensing range from the portable spectro-radiometer, which measures all wavelengths between 0.4 and 1.1  $\mu\text{m}$  to radiometers with multiple channels set for discrete wavebands. Instruments with individual channels mimic the wavebands available on the current satellites. A rapidly emerging technology that has yet to be applied is the use of multiple waveband video cameras. This system offers a capability not possible with other radiometers, in that the data are readily available for viewing without intense signal processing and manipulation. Video camera systems may provide a practical tool for disease assessment.

### **Vegetative Indices**

To use the information contained in the reflectance across wavelengths, several vegetative indices have been proposed and evaluated. These indices are based primarily on the ratio or difference between the reflectance in the near-infrared and red wavelengths. The approaches range from simple ratios of near-infrared/(near-infrared+red) is more appropriately related to the interception of photosynthetically active radiation. To account for the soil background the perpendicular vegetative index was developed to account for a changing soil background due to surface soil water content changes. There have been several other indices developed to describe how the changing reflective properties change with growth of the plant.

Observed changes in the vegetative index, in particular, the ratio vegetative index and the normalized vegetative index have shown unique seasonal patterns. The patterns of both indices show an increase with the developing canopy and a hysteresis effect during senescence because plant material remains standing in the field, which has different reflective properties than the soil in the background. Over fields of seemingly uniform conditions there is considerable variation in the observed signal whether the data are collected with hand-held, boom-mounted, or aircraft-mounted systems. The variation is typically 10% of the field mean. However, the change in spatial variability may be one of the methods that could be effectively used to monitor the changes that occur within fields as a result of disease. Most diseases do not infect a whole field uniformly and thus could induce a change in the field pattern. Even on a single sample event this method could provide valuable information given a priori knowledge about the expected level of field variability.

### **Emitted Radiation**

All objects that have a temperature emit radiation according to Planck's Law. Soil and plant canopies emit energy, and given the temperatures found on the earth's surface, range in the 10-14 **µm** waveband. Temperature of a plant canopy can be described by the temperature of individual leaves, the temperature of foliage, or the temperature of the canopy that includes the soil. Leaf temperatures that have been measured relative to the occurrence of Verticillium wilt or brown rot of soybeans (*Phialophora gregata* Gans) have been measured with attached leaf thermocouples. Other measurements of Verticillium wilt have been made with infrared thermometers. Each method has provided a unique relationship of describing the change in leaf temperature relative to the presence of disease.

### **Energy Exchange Processes**

Temperatures of the leaf, foliage, or canopy are a result of the energy exchange process. The observed temperatures are a result of the partitioning between the sensible and latent heat exchanges and therefore are a balance between the energy impinging on the leaf or foliage and the water available for evaporation. Simply stated then, a surface with a free water surface will be as cool as possible given the meteorological conditions while one without water will be the warmest possible under a given set of conditions. It is this relationship that has allowed foliage temperature to be effectively used in the estimation of transpiration from canopies. Also, any factor that disrupts this water flow to the leaf, e.g., vascular diseases, root diseases, or diseases

that disrupt the stomatal action, will cause the foliage temperature to be higher than that observed in healthy foliage. We have been successful in using foliage and canopy temperature in evapotranspiration models for a variety of crops. In well-irrigated crop canopies, the variation across a field is relatively uniform and the variation increases with increasing soil water deficits. As with the reflected radiation, the change in field variability may be useful in defining the characteristics of a given field.

### **Crop Stress Indices**

To improve the efficiency of using foliage temperatures, several crop water stress indices have been proposed and evaluated since the middle 1970s. These became possible at this time due to the leaf area index, while the normalized difference [(near-infrared development of the accurate, portable, hand-held infrared thermometer). At first, the comparison was made between the foliage and air temperature ( $T_f - T_a$ ), since this form was the integral part of the energy exchange process. It was found that although the  $T_f - T_a$  differences were related to crop yield induced by water stress, the relationships were site dependent. Further development and study revealed that other environmental variables were needed to fully interpret foliage temperatures and develop less site-specific relationships. The primary variables were net radiation, wind speed, and vapor pressure deficit. These stress indices have been based on the energy exchange principles between the foliage and the surrounding atmosphere. Any factor that affects the rate of water movement to the leaf has an impact on the foliage temperature. For example, the addition of high salt content irrigation even in large amounts causes the foliage temperature to be warmer than those plants irrigated with the same volume of salt-free water.

Recent research has identified that plants have biochemical temperature optima that define the optimum temperature for plant growth. Combining foliage temperature with these predetermined optimum temperatures provides another description of plant stress. It has been found that plants with maximum growth in a particular environment have the minimum amount of time outside of this predetermined thermal range. For cotton, this range was determined to be 23-30 C and for wheat 18-25 C. This range has been defined as the thermal kinetic window and is based on the biochemical efficiency of a particular plant. The utility of this stress index has yet to be fully evaluated; however, it offers a method of linking the plant response to an observed parameter. Eg: foliage temperature.

Other methods that can be used are to calculate the canopy resistance to water vapour exchange. It is known that many diseases affect the stomatal resistance and the combination of energy balance and observed foliage temperature provide a method of estimating canopy resistance. These techniques, however, are yet to be applied to any measure of disease. They may offer the potential of quantifying the degree of stress or level of infection in ways that have not been possible before.

The instruments available for measuring the foliage temperature range from hand-held portable units, to fixed, battery-powered systems to airborne or satellite thermal scanner systems. The coverage is obtained, e.g., NOAA or GOES satellites. Smaller areas are covered with systems that provide less frequent coverage, e.g., **LANDSAT** or **SPOT**. The variation within a scene of data obtained with an airborne or satellite system will not permit the reliable detection of the onset of a disease. The handheld or fixed units on the ground may be useful in a research setting to determine the causal relationships and the development of a monitoring program where a problem is suspected. Each of these instruments require some training to most properly collect and interpret the data.

### **Temporal and Spatial Variation**

Both the temporal and spatial attributes of remote sensing techniques detect unique features about the surface being observed. Satellites that provide repeated coverage of the earth have allowed assessments to be made of the changes that have occurred over a period of months or years. The same factors apply when applying remote sensing to the monitoring of agricultural fields, forests, and native or managed grasslands. The value of repeated coverage has provided for a unique glimpse at the ecosystem that we are trying to monitor.

Temporal variations can be large because the system to which we are applying may be changing due to the normal progression of growth. However, we know what patterns to expect and deviations away from that pattern provide an investigative tool. Likewise, changes in the spatial patterns may signal a potential problem within a given field or ecosystem. The interpretation of a temporal and spatial pattern will require some experience but may provide an indication of a problem not possible before this information was made available.

### **Integrating Remote Sensing Into Plant Pathology**

There are two avenues in which remote sensing information, either reflected or emitted radiation, can be incorporated into disease monitoring. The two approaches involve either direct



or indirect methods of evaluating the disease occurrence and extent. Given the number of factors that cause variation in both the reflected and emitted radiation signals it is unlikely that the direct monitoring method will be useful. Both the indirect and direct methods require a priori knowledge that a condition may exist.

Given this knowledge then, one may use a direct monitoring program to measure the extent of a disease. eg: *Phytophthora* spp. on soybeans or *Fusarium* spp. on beans, which cause a reduction in leaf area. The spatial sampling capability provides an assessment not possible with ground monitoring.

The indirect method would involve interpretation of deviations from the expected case either in temporal or spatial patterns. For example, an increase in foliage temperature in a field with an adequate soil water supply would signal a potential problem that could invoke a monitoring effort. An unexplained change in leaf area or wilting resulting in a change in reflectance could signal a problem before complete infestation. The utilization of both indirect and direct methods will require imagination and dedication to the problem by a number of researchers.

## **Lecture 04**

### **General principles of plant diseases management – Importance, general Principles – Avoidance, exclusion, eradication, protection and therapy, immunization**

Information on etiology, symptoms, pathogenesis and epidemiology of plant diseases are intellectually interesting and scientifically justified but most important of all they are useful as they help in formulation of methods developed for successful management of disease and thereby increasing the quantity and improving the quality of plant and plant products. Practices of disease management vary considerably from one disease to another depending upon the type of pathogen, the host and the biotic and abiotic factors involved. Contrary to management of human and animal diseases where every individual is attended, the plants are generally treated as populations and measures used as preventive rather than curative.

Methods for plant diseases control were first classified by Whetzel (1929) into exclusion, eradication, protection and immunization. Further advances in plant pathology leading to development of newer methods. Two more principles - avoidance and therapy were created (NAS, 1968)

#### **Avoidance**

It involves avoiding disease by planting at time when, or in areas where inoculum is absent or ineffective due to environmental conditions. The major aim is to enable the host to avoid contact with the pathogen or to ensure that the susceptible stage of the plant does not coincide with favourable conditions for the pathogen. The main practices under avoidance are choice of geographical area, selection of the field, choice of sowing/ planting time, selection of seed and planting material, short duration/ disease escaping varieties and modification of agronomic/ cultural practices. The potato cultivation at high altitude is relatively free from viruses; as prevailing environmental conditions do not permit the buildup of vector populations. Similarly, early planting of potato or wheat, in Indo Gangetic plains may escape late blight or stem rust damage respectively.

#### **Exclusion**

It means preventing the inoculum from entering or establishing in a field or area where it does not exist. Seed certification, crop inspection, eradication of inoculum and / or insect vectors, and quarantine measures are some of the means of preventing the spread of pathogens.

### **Eradication**

The process of reducing, inactivating, eliminating or destroying inoculums at the source, either from a region or from an individual plant in which it is already established is termed as eradication. Eradication involves eliminating the pathogen from infested areas; the magnitude of the operation involved may vary considerably. One of the most extensive eradication operations carried out so far was to get rid of the citrus canker (*Xanthomonas axonopodis*) in the USA during 1927-35. As many as 4 million citrus trees were cut and burnt at a cost of about 2.5 million dollars to eradicate the pathogen. The practices invariably employed to achieve eradication of inoculums include eradication of alternate and/or collateral hosts, crop rotations, field sanitation, heat or chemical treatments of plant materials or soil, biological control etc.

### **Protection**

The protection of infection courts against the inoculums of many fast spreading infectious pathogens, brought by wind from neighboring fields or any other distant place of survival. Principles of avoidance, exclusion and eradication may not be sufficient to prevent the contact of host with pathogen, thus development of the disease is imminent. Measures are necessary to protect host plants from invading inoculums. It can be achieved by creating toxic barrier between the plant surface and the inoculums. Methods employed to achieve such results are chemical sprays, dusts, modification of environment, and modification of host nutrition.

### **Host resistance**

It utilizes in-built mechanism to resist various activities of pathogen. The infection or subsequent damage by pathogen can be rendered ineffective through genetic manipulation or by chemotherapy. The host resistance can also be induced by use of certain biotic and abiotic factors. The discovery of Mendelian laws of inheritance and developments in plant breeding techniques have helped in developing crop varieties resistant to specific pathogen or group of pathogens. The classical breeding techniques include selection, mutation and hybridization. Use of biotechnological tools such as tissue culture, genetic engineering and protoplast fusion are being used to develop resistant cultivars of various economically important crops.

### **Therapy**

It is the treatment of infected host plant, which is attempted in case of economically important horticulture plants. As a principle of plant disease control, it provides an

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opportunity

to cure or rejuvenate the diseased host plant by use of physical or chemical agents. The first five of

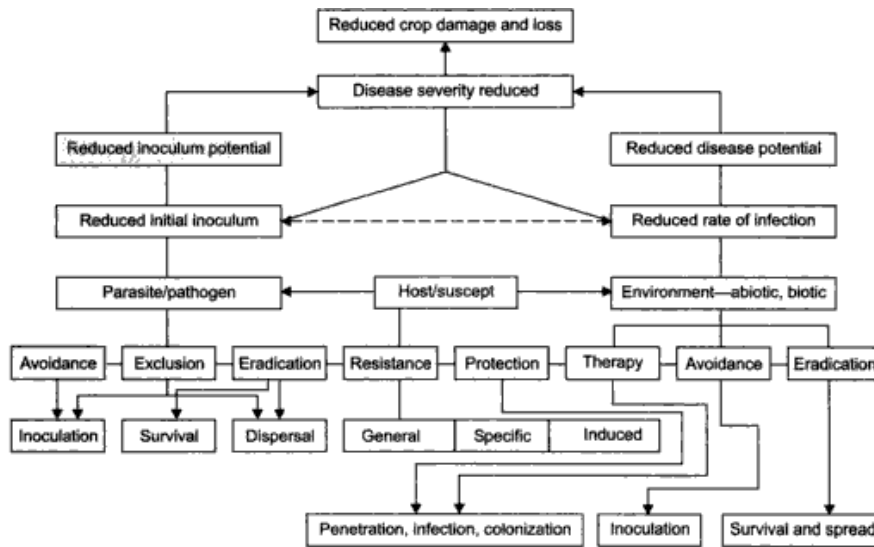
these principles are mainly preventive (prophylactic) and constitute the major components of plant disease management. They are applied to the population of plants before infection takes place. Therapy is a curative procedure and is applied to individuals after infection has taken place. Under the concept of disease management these principles have been classified into following five categories:

1. Management of physical environment (cultural control)
2. Management of associated microbiota (biological antagonism)
3. Management of host genes (host resistance)
4. Management with chemicals (Chemical control)
5. Management with therapy (Physical, chemical etc)

These six principles that characterize the modern concept of plant disease management should be viewed from three standpoints

- (a) Reduction in the initial inoculum or the rate of disease development.
- (b) Management of the pathogen population, the cure or induced defense of the susceptible or modify the environment as it influences disease and
- (c) Interruption of dispersal, survival or the course of disease development.

These interactions are originally proposed by Baker (1968) and Roberts and Boothroyd (1972) and subsequently modified for the readers are illustrated as below:

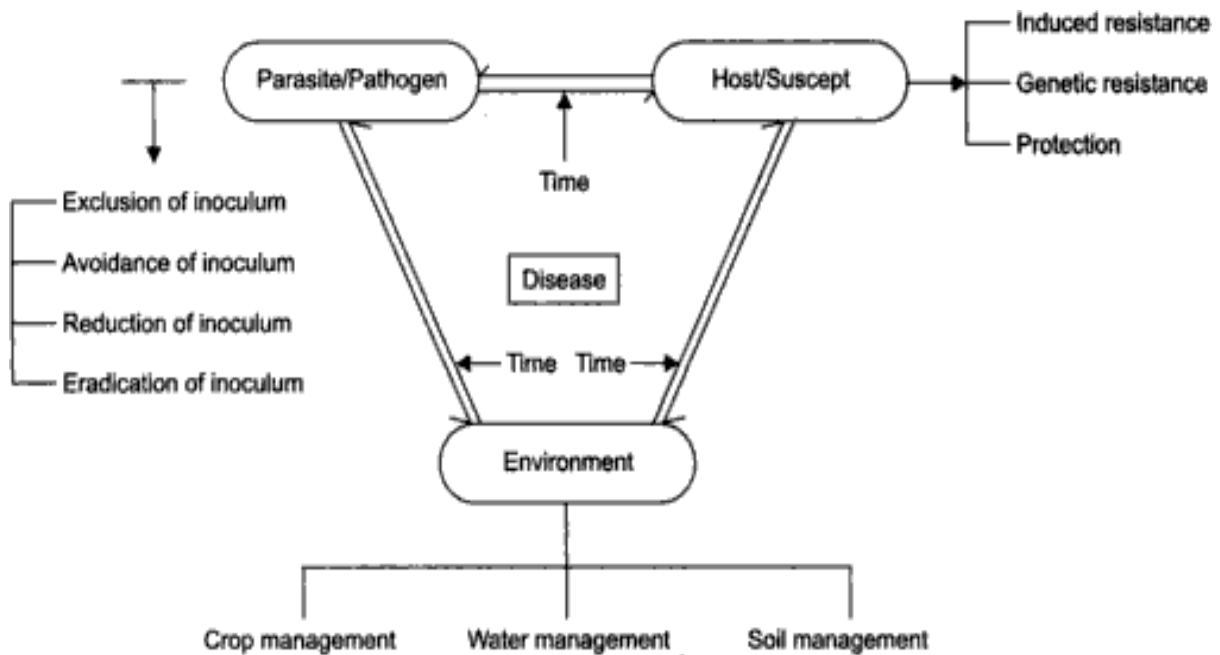


### Integrated disease management

The term Integrated pest management was originally designed for management of insect pest but it is equally applicable to plant diseases also. IPM is an ecosystem- based strategy that focuses on long term prevention of pests or their damage through a combination of techniques such as biological control, habitat manipulation, and modification of cultural practices and use of resistant varieties.

Management of pathogen involves the practices directed to exclude, reduce or eradicate inoculum.

Management of the host involves the practices directed to improve plant vigor and induce resistance through nutrition, introduction of genetic resistance through breeding and providing need based protection by chemical means. Management of environment involves the practices that modify the environment which is not favorable to pathogen or disease development and does not predispose host to attack.



Lecture 05

**Regulatory methods – Plant Quarantine and Inspection – Quarantine Rules and Regulations**

**Plant Quarantine**

The term ‘Quarantine’ means simply forty i.e., 40 days period. This was more commonly referred to the period of detention for ships arriving from countries subject to epidemic diseases such as the Bubonic plague, cholera and yellow fever. The crew and the passengers used to be compelled to remain isolated on board for sufficient period to permit the diseases to develop and be detected. The purpose of the health authorities was to establish adequate detention period. Later on, the term ‘Quarantine’ came to be only used for the detention and the practices connected with it. The term got associated from the human disease field to the animal disease field and later on adopted to cover protective methods for the exclusion of pests and diseases of agricultural and horticultural crops.

In strict sense ‘Plant Quarantine’ refers to the holding of plants in isolation until they are believed to be healthy. Now, broader meaning of the plant quarantine covers all aspects of the regulation of the movement of living plants, living plant parts/plant products between politically defined territories or ecologically distinct parts of them. Intermediate quarantine and post entry quarantine are used respectively to denote the detention of plants in isolation for inspection during or after arrival at their final destination.

**Importance**

The entry of a single exotic insect or disease and its establishment in the new environment continues to cause great, national loss (table ) till such time it is brought under effective control. In certain cases a country has to spend a few million rupees before success in controlling the introduced insect pest or disease is achieved.

**Losses caused by introduced plant diseases**

Disease	Host	Country	Introduced from	Losses caused
Canker	Citrus	U.S.A	Japan	\$13 million; 19.5 million trees destroyed
Dutch elm	Elm	U.S.A.	Holland	\$25 million-\$ 50,000 disease million

Blight	Chestnut	U.S.A.	Eastern Asia	\$100-1000 million
Powdery mildew	Grapevine	France	U.S.A	80% in wine production
Downy mildew	Grapevine	France	U.S.A	\$ 50,000 million
Bunchy top	Banana	India	Sri Lanka	Rs. 4 crores
Wart	Potato	India	Netherlands	2500 acres infected
South American leaf blight	Rubber	Dutch– Brazil	Guiana	40,000 trees destroyed
do	-do-	North Columbia	Brazil	78% trees destroyed
Blue mould	Tobacco	Europe	U.K	.\$ 50 million
–do	--do-	Sweden	U.K.	1.2 million Kroner

### **History**

The first plant quarantine law was promulgated in Rollen, France in 1860 to suppress and prevent the spread of common barberry, the alternate host for wheat stem rust. Among other countries, the first few to establish plant quarantine services were Germany, France, Australia and the U.S.A. In India, legislative measures against crop pests and diseases was initiated under the Destructive Insects and Pests Act of 1914 (DIP Act) and it was passed by Governor General of India on 3<sup>rd</sup> February, 1914. Under this Act, rules governing the import and movement of plants and plant materials, insects and fungi are framed. The Act provides

- It authorizes the Central Government to prohibit or regulate the import into India or any part thereof any specific place therein, of any article of class of articles.
- It authorizes the officers of the Customs at every port to operate, as if the rules under the D.I.P. Act made under the Sea Customs Act.

1. It authorizes the Central Government to prohibit or regulate the export from a State of the transport from one State to another State in India of any plants and plant materials, diseases or insects likely to cause or infestation. It also authorizes the control of transport and carriage and



gives power to prescribe the nature of documents to accompany such plants and plant materials and articles.

2. It authorizes the State Governments to make rules for the detention, inspection, disinfection or destruction of any insect or class of insects or of any article or class of articles, in respect of which the Central Government have issued notifications. It also authorizes the State governments for regulating the powers and duties of the officers whom it may appoint on this behalf.

3. It provides penalty for persons who knowingly contravene the rules and regulations issued under the Act.

4. It also protects the persons from any suit or prosecution or other legal proceedings for anything done in good faith or intended to be done under the Act. Consequent to Bengal famine 1943, a Central Plant Protection organization was established in 1946 under the then Ministry of Food and Agriculture. Often a new pest, disease or weed has accidentally entered a country where it did not exist before and has multiplied, spread and caused enormous damage to the crops of that country.

For instance powdery mildew of grapevine (*Plasmopara viticola*), introduced into France from America, was responsible for the destruction of the vine industry of that country until hybridization with resistant American stock offered a solution. The blight disease of chestnut (*Endothia parasitica*) which was introduced into U.S.A. from Asia in 1904, completely wiped out chestnut trees. Coffee rust (*Hemileia vastarix*) which came into India in 1879 from Sri Lanka is now widespread in all coffee growing areas. Fire blight (*Erwinia amylovora*) of pear and other pomes which was introduced from England in 1940 is well established in Uttar Pradesh. Late blight (*Phytophthora infestans*) of potato introduced into India in 1889 from Europe is now present in many parts of the country. Flag smut (*Urocystis tritici*) of wheat introduced from Australia is now well spread in Madhya Pradesh, Punjab, Rajasthan and Uttar Pradesh. Rubber powdery mildew (*Oidium heavea*), which was introduced from Malaysia in 1938, is also causing great concern in Kerala. Black rot of crucifers (*Xanthomonas campestris* pv. *campestris*) believed to have been introduced to India with seeds imported from Holland, and other European countries after World War II, prevailed for some years on the hills and then spread to the plains and became established in Indian seed stocks, especially in West Bengal. Among the more important plant disease introductions, mention may be made of bunchy top virus of banana introduced from Sri Lanka in 1940 which has since spread widely in Kerala, Orissa, West Bengal

and Assam. The wart disease (*Synchytrium endobioticum*) of potato was first noticed in Darjeeling district of West Bengal having been introduced with seed potatoes from Holland. By 1962, the disease spread over nearly 1000 ha and has recently been reported from Nepal also. The mosaic disease of banana is another introduced disease which is only confined to Gujarat and Maharashtra states. Recently the apple scab (*Venturia inaequalis*) which was only confined to a small area in Jammu and Kashmir has now appeared in severe form in many locations in Himachal Pradesh, and is posing a problem to the apple industry. The establishment of a plant quarantine regulation should rest on the following fundamental pre-requisites.

- i. The pest/disease under consideration must be one that will offer actual or expected threats to substantial interests (Agricultural and / or commercial)
- ii. The quarantine regulation or degree must represent a measure for which no substitute action involving less interference with normal activities is available.

**Diseases believed to have been introduced into India from foreign countries**

Disease	Host	Date of first record	Introduction from
Leaf rust ( <i>Hemileia vastarix</i> )	Coffee	1879	Sri Lanka
Late blight ( <i>Phytophthora infestans</i> )	Potato	Tomato 1883	Europe
Rust ( <i>Puccinia carthami</i> )	Chrysanthemum	1904	Japan or Europe
Flag smut ( <i>Urocystis tritici</i> )	Wheat	1906	Australia
Downy mildew ( <i>Plasmopara viticola</i> )	Grapevine	1910	Europe
Downy	Cucurbits	1910	Sri Lanka

mildew( <i>Pseudoperonospora cubensis</i> )			
Downymildew( <i>Sclerospora philippinensis</i> )	Maize	1912	Java
Foot rot ( <i>Fusarium moniliforme</i> var. <i>majus</i> )	Rice	1930	SouthEast Asia
Leaf spot( <i>Phyllachora sorghi</i> )	Sorghum	1934	SouthAfrica
Powdery mildew( <i>Oidium heveae</i> )	Rubber	1938	Malaya
Blackshank( <i>Phytophthora parasitica</i> var. <i>nicotianae</i> )	Tobacco	1938	DutchEast Indies
Fire blight Pear and other( <i>Erwinia amylovora</i> )	pomes	1940	England
Crown-gall and hairy root ( <i>Agrobacterium tumefaciens</i> ssp. <i>rhizogenes</i> )	Apple, Pear	1940	England

1. Bunchy top Banana 1940 Sri Lanka
2. Canker Apple 1943 Australia (*Sphaeropsis malorum*)
3. Wart Potato 1953 Netherlands (*Synchytrium endobioticum*)

Despite every precaution of inspection, certification and treatment, it is not always possible to guarantee that a consignment is completely free from pathogens. In doubtful cases it is advisable to subject plants to a period of growth in isolation under strict supervision in the importing country (post-entry quarantine). The plants are grown at a quarantine station. When direct importation of plants to a country's own quarantine station is considered very dangerous, quarantine during transit from the country of origin (intermediate quarantine) may be required.

The requirements of an intermediate station are similar to those for a post-entry station. Intermediate quarantine inspection must always be followed by post-entry quarantine after arrival of the consignment at its final destination. During post-entry or intermediate quarantine plants must be kept under close supervision, so that any pest or disease which appears may be immediately detected and grown under optimum conditions, so that symptoms are not marked by physiological disturbances.

International plant protection convention the first effort towards international agreement on Plant Protection was made in 1914 under the auspices of the International Institute of Agriculture in Rome. This was followed by an International Convention of Plant Protection by over 50 member countries of the Institute in 1919 and certain Agreements regarding the issue and acceptance of phytosanitary certificates were finalized. The project received a set back due to Second World War and was later on revived by the FAO. In post-war period International action in Plant Protection and particularly in plant quarantine was encouraged by FAO with the establishment in 1951 of the International Plant Protection Convention. This agreement was constituted with the purpose of securing common and effective action to prevent the introduction and spread of pests and diseases of plants and plant products as to encourage Governments to take all steps necessary to implement its prevention (Ling, 1953).

The following regional Plant Protection Organizations are now in operation.

1. The European and Mediterranean Plant Protection Organization (EPPO)
2. The Inter-African Phytosanitary Council (IAPSC)
3. Organismo Internacional Regional de Sanidad Agropecuaria (OIRSA)
4. The Plant Protection Committee for the South East Asia and Pacific region.
5. Comité Interamericano de Protección Agrícola (CIPA)
6. The Caribbean Plant Protection Commission (CPPC)
7. The North American Plant Protection Organization (NAPPO).

Under article 3 of that International Plant Protection Convention, the Plant Protection Agreement for South East Asia and Pacific Region was sponsored by F.A.O in 1956, and India became a party to this Agreement in the same year along with Australia, Sri Lanka, the U.K., Laos, Netherlands, Indonesia, Portugal and Vietnam. Our Government agreed to adopt

legislative measures specified in the Convention for the purpose of securing common and effective action to prevent the introduction and spread of pests and diseases of plants and plant products and to promote measures for their control and also agreed to assume all responsibilities for the fulfillment within its territories of all requirements under the Convention. It was agreed that the Government shall make provision for:

a. An official plant protection organization, with the following main functions:

1. The inspection of growing plants, of areas under cultivation and of plants and plant products in storage and in transportation with the object of reporting the existence, outbreak and spread of plant diseases and pests and of controlling those pests and diseases.

2. The inspection of consignments of plants and plant products moving in international traffic, the inspection of consignments of other articles or commodities moving in international traffic under conditions where they may act incidentally as carriers of pests and diseases of plants and plant products and the inspection and supervision of storage and transportation facilities of all kinds involved in international traffic whether of plants and plant products or other commodities, with the object of preventing the dissemination across national boundaries of pests and diseases of plants and plant products.

3. The disinfection or disinfection of consignments of plants and plant products moving in international traffic, and their containers, storage places, or transportation facilities of all kinds employed.

4. The issue of certificates relating to phytosanitary condition and origin of consignments of plants and plant products (Phytosanitary certificates).

b. The distribution of information within the country regarding the pests and diseases of plants and plant products and the means of their prevention and control

c. Research and investigation in the field of plant protection. A revised text of convention was approved in 1979. As of December 1980, the number of states party to the convention is 81. Besides this world-wide convention, other regional agreements and organizations have been created to safeguard the interests of groups of neighbouring countries with similar plant protection problems.

Regional action is needed to prevent a pathogen or pest absent from a whole area from being introduced into any part of the area, as its entry into one territory will endanger neighbouring countries.

## **Plant quarantine methods**

There are number of plant quarantine methods which are used separately or collectively to prevent or retard the introduction and establishment of exotic pests and pathogens. The components of plant quarantine activities are:

### **1. Complete embargoes**

It involves absolute prohibition or exclusion of specified plants and plant products from a country infected or infested with highly destructive pests or diseases that could be transmitted by the plant or plant products under consideration and against which no effective plant quarantine treatment can be applied or is not available for application.

### **2. Partial embargoes**

Partial embargoes, applying when a pest or disease of quarantine importance to an importing country is known to occur only in well defined area of the exporting country and an effectively operating internal plant quarantine service exists that is able to contain the pest or disease within this area.

### **3. Inspection and treatment at point of origin**

It involves the inspection and treatment of a given commodity when it originates from a country where pest/disease of quarantine importance to importing country is known to occur.

### **4. Inspection and certification at point of origin**

It involves pre-shipment inspection by the importing country in cooperation with exporting country and certification in accordance with quarantine requirements of importing country.

### **5. Inspection at the point of entry**

It involves inspection of plant material immediately upon arrival at the prescribed port of entry and if necessary subject to treatment before the same related.

### **6. Utilization of post entry plant quarantine facilities**

It involves growing of introduced plant propagating material under isolated or confined conditions.

## **Plant quarantine organizations in India**

The first recorded plant quarantine measure in India dates back to 1906 when perceiving the danger of introducing the Mexican boll weevil, the Government of India directed that all cotton imported from the New World should only be admitted to India after fumigation with

carbon disulphide at the port of entry. In India two categories of regulatory measures are in operation for controlling pests, diseases and weeds. In the first category regulatory measures are aimed to prevent the introduction of exotic pests and diseases into the country or their spread from one State or Union Territory to another (Plant Quarantine).

The second pertains to suppression or prevention of spread of pests and diseases in localized areas within a State or Union Territory. The former derives its authority from the Destructive Insects and Pests (DIP) Act 1914 of the Central Government and the latter from Agricultural Pests and Diseases Acts of the various States. The legislative measures against crop pests and diseases were initiated under the DIP Act of 1914 which was passed by the then Governor General of India in Council on 3 February 1914. Prior to the establishment of the Directorate of Plant Protection, Quarantine and Storage in 1946, under the Ministry of Food and Agriculture, the various rules and regulations of the DIP Act were enforced by the customs department. The quarantine regulations are operative through The Destructive Insects and Pests Act, 1914 (which has been revised 8 times from 1930 to 1956 and amended in 1967 and 1992).

The provisions of the DIP Act are

1. It authorizes the Central Government to prohibit or regulate the import into India or any part thereof for any specific place therein of any article or class of articles.
2. It authorizes the officers of the Customs at every port to operate, as if the rules under DIP Act are made under the Sea Customs Act.
3. It authorizes the Central Government to prohibit or regulate the export from a State or the transport from one State to another State in India of any plants and plant material, diseases or insects, likely to cause infection or infestation. It also authorizes the control of transport and carriage and gives power to prescribe the nature of documents to accompany such plants and plant materials and articles.
4. It authorizes the State Governments to make rules for the detention, inspection, disinfection or destruction of any insect or class of insects or any article or class of articles, in respect of which the Central Government has issued notification. It also authorizes the State Governments for regulating the powers and duties of the officers whom it may appoint on its behalf.
5. It provides penalty for persons who knowingly contravene the rules and regulations issued under the Act.

6. It also protects the personnel from any suit or prosecution or other legal proceedings for anything done in good faith as intended to be done under this Act.

The quarantine regulations are operative through "The Destructive Insects and Pests Act, 1914 (which has been revised and time from 1930 to 1956 and amended in 1967 and 1992. The Act also empowers the State Governments to frame suitable rules and issue notifications for inter-state movement of plant and plant material. Those rules are known as plant quarantine rules. Under the Act, Central Government frames rules prescribing the seaports, airports and land frontiers through which plants and specified plant material can enter India, and the manner in which these can be imported. The DIP Act operates under the National Sea Customs Act and the points of entry are located within the jurisdiction of State on the advice of Central Government, the State frames rules for detention, inspection, disinfection and destruction (as against entry) of material, if required, and delegates powers in this regard to concerned authorities with the enforcement of rules.

The plant quarantine service is centrally organized and administered through the Directorate of Plant Protection, Quarantine and Storage established under the Ministry of Agriculture (Department of Agriculture and Co-operation) which is headed by the Plant Protection Adviser to the Government of India and having its headquarters at N.H. IV, Faridabad, Haryana State. Import regulations When plants are imported the following principles should be followed. Some plant pathogens and pests are generally distributed in most parts of the world but others are more or less restricted in their occurrence.

In some cases this limitation is due to such factors as unsuitable environmental conditions or lack of the required host plant, but in many other cases the absence of a pathogen. Most countries are aware of the desirability of delaying for as long as possible the arrival of exotic pathogens and take action to prevent their spread by introducing legislation and setting up organizations to prevent their entry. Plant quarantine legislation varies from country to country but in most cases it restricts or prohibits the importation of the pests or pathogens themselves, plants on which they might be living, soil which might be infested, foodstuffs which might carry them, and packing materials, particularly those of plant origin. Good legislation is as brief and clear as possible, at the same time being easy to interpret, gives adequate protection without interfering more than is essential with trade, and contains only restrictions which are



scientifically justifiable. When plants are imported there are certain principles which, if followed ensure that as few risks as possible are taken.

1. Import from a country where, for the crop in question, pathogens which are particularly to be guarded against are absent.
2. Import from a country with an efficient plant quarantine service, so that inspection and treatment of planting material before despatch will be thorough, thus reducing the likelihood of contaminated plants being received.
3. Obtain planting material from the safest known source within the selected country.
4. Obtain an official certificate of freedom from pests and diseases from the exporting country. Treatment of the material in the country of origin may be done; this should be noted on the certificate.
5. The smaller the amount the less the chance of its carrying infection, and inspection as well as post-entry quarantine.
6. Inspect material carefully on arrival and treat (dust, spray, fumigate, heat treat) as necessary.
7. Import the safest type of planting material, e.g. seeds are usually safer than vegetative material, unrooted cuttings than rooted. The use of axenic cultures of meristem tip tissues (micropropagation) for the international exchange of germplasm material has outstanding advantages, as such tissues can be expected to be free from latent infections by viruses, phytoplasmas etc., as well as other pathogens which are more readily detectable by visual means.
8. If other precautions are not thought to be adequate, the consignment for import should be subject to intermediate or post-entry quarantine. Such quarantine must be carried out at a properly equipped station with suitably trained staff.

Seed was not originally included in the DIP Act, but because of the changing situation and to meet the current requirements, the Government of India passed the Plants, Fruits, Seeds (Regulation of Import into India) Order 1984 which came into effect in June 1985. The conditions for the import of 17 crops are stipulated in this order. The main features of the order are:

1. Seed has been brought under the purview of the DIP Act.
2. No consignment can be imported into the country without valid import permit issued by the Plant Protection Adviser to the Government of India.

3. No consignment can be imported without an official phytosanitary certificate issued by the plant quarantine agency of the exporting country.

4. Post-entry growth of the specified crops at approved locations.

### **A. Conditions for import**

In India, there are general and specific conditions for the import of plants (including bulbs, tubers, rhizomes, corms, cuttings, buddings, grafts, layers, suckers, roots and flowers) and plant materials (including plant products such as ginned cotton, unmanufactured tobacco etc.).

### **General conditions**

1. Import permits are essential for:

- a. Seeds and fruits for consumption,
- b. Seeds and plants for sowing or planting,
- c. Soil, earth, clay for microbiological, soil mechanics or mineralogical investigations
- d. Peat for horticultural purposes
- e. Live insects and f. Living fungi in pure culture, including *Rhizobium* cultures.

2. All plants should be accompanied by Phytosanitary certificate from the country of origin.

3. All plants on arrival at port, shall be inspected and if necessary fumigated, disinfested or disinfested by Plant Protection Adviser to the Government of India or any other officer authorized by him on his behalf.

4. Plants and seeds which require post-entry quarantine inspection shall be grown in post-entry quarantine facilities approved by the Plant Protection Adviser to the Government of India.

5. Import of hay or straw or any material of plant origin used for packing is prohibited.

6. Import of soil, earth, compost, sand, plant debris along with plants, fruits and seeds is prohibited.

**Note:** Cut flowers, garlands, bouquets, fruits and vegetables weighing less than 2 kg for personal use may be imported without a permit or phytosanitary certificate, but are subject to inspection. Special conditions In addition to the general conditions, there are special conditions for certain notified plants as follows.

**1. Prohibition from certain areas**

Name of the plant	Countries from where prohibited
Cocoa and all species of Sterculiaceae and Bombaceae	Africa, Sri Lanka, West Indies and
Coffee beans	Africa, South America, Sri Lanka
Rubber	South America, West Indies
Sugarcane	Australia, Fiji, Papua New Guinea
Sunflower	Argentina, Peru

1. Prohibited for general public: Coconut plants and seeds, coffee plants and seeds, cotton seeds and unginning cotton, forest tree seed (*Castanea*, *Pinus*, *Ulmus*), groundnut seeds and cuttings, potato, sugarcane, tobacco seeds and wheat seeds.

2. Plants/seeds which require post entry quarantine: Cocoa, citrus, coconut, groundnut, potato, sugarcane, sunflower, tobacco and wheat.

3. Additional declarations required for notified plants (see Table below)

**Plant/seed Additional declarations for freedom of pests**

All species of <i>Allium</i> (onion, garlic, leek, chive, shallot, etc.).	Smut ( <i>Urocystis cepulae</i> )
Cocoa and all of the family species Sterculiaceae and Bombaceae	Podrot ( <i>Monilia rorei</i> ), Mealy pod ( <i>Trachysphaeria and fructigena</i> ), Witches' broom ( <i>Crinipellis perniciosus</i> ) Swollen shoot Virus
All species of <i>Citrus</i> (lemon, lime, orange etc.,)	Mal secco ( <i>Deuterophoma tracheiphila</i> )
Coconut seeds and all species of <i>Cocos</i>	Lethal yellowing, Cadang, Bronze leaf wilt, Guam, Coconut disease, Leaf scorch
Coffee plants, seeds	American leaf spot ( <i>Omphalidiflvida</i> ), virus diseases

Cottonseeds	Bacterialblight( <i>Xanthomonasaxonopodis</i> pv. <i>malvacearum</i> and <i>Glomerellagossypii</i> )
Forestreeseeds(allspecies <i>Cronartiumribicola</i> , <i>E ndothea</i> of <i>Pinus</i> , <i>Ulmus</i> , <i>Castanea</i> )	<i>parasitica</i> , <i>Ceratocystisulmi</i> , <i>Dothiostromapini</i> .
Groundnut seeds (all speciesof <i>Arachis</i> )	i. production of seeds in areas free of <i>Pucciniaarachidis</i> and <i>Sphaceloma arachidis</i> . ii. Inspection of parent crops in active growingseasonsandcertificationforfreedomfro mpeanutmottle,peanutstunt,marginalchlorosis andpeanut stripeviruses
Lucerne(all speciesof <i>Medicago</i> )	Bacterialwilt( <i>Corynebacterium incidiosum</i> )
Potato(allspeciesof <i>Solanum</i> )	Wart( <i>Synchytriumendobioticum</i> )andfreedom ofparentcropfromvirusdiseases
Rubber(allspeciesof <i>Hevea</i> )	SouthAmericanleafblight( <i>Microcyclusulei</i> , <i>Sphaerostilberegens</i> )
Sugarcane(allspeciesof <i>Saccharum</i> )	Leafscald( <i>Xanthomonasalbineans</i> ),Gummosis ( <i>Xanthomonas vasculorum</i> ), Sereh,downymildew,chloroticstreakandFiji disease.

**Agenciesinvolvedin plantquarantine**

The authority to implement the quarantine rules and regulations framed under DIP Act rests basically with the Directorate of plant Protection, Quarantine & Storage, under the Ministry of Agriculture. This organization handles bulk import and export of seed and planting material for commercial purpose. Under this organization 9 seaports, 10 airports and 7 land frontiers are functioning. These are the recognized ports for entries for import of plant and plant material. The names and places of the ports and stations are as follows.

**A. Seaports-Place/State/Union territory**

1. Bhavnagar-Gujarat
2. Calcutta-West Bengal
3. Chennai-TamilNadu
4. Cochin-Kerala
5. Mumbai-Maharashtra
6. Nagapattinam-TamilNadu
7. Rameswaram-TamilNadu
8. Tuticorin-TamilNadu
9. Visakhapatnam- AndhraPradesh

**B. Airports**

1. Amritsar-Punjab
2. Calcutta-West Bengal
3. Chennai-TamilNadu
4. Hyderabad-AndhraPradesh
5. Mumbai-Maharashtra
6. NewDelhi- NewDelhi
7. Patna-Bihar
8. Tiruchirappalli-TamilNadu
9. Trivandrum-Kerala
10. Varanasi -UttarPradesh

**C. Landfrontiers**

1. AmritsarRailwayStation-Punjab
2. AttariRailwayStation-Punjab
3. Attari-WagahBorder-Punjab
4. BangaonBenapolBorder-West Bengal
5. GedeRoad RailwayStation-West Bengal
6. Kalimpong-WestBengal
7. SukhiaPokhri -WestBengal

The Government of India has also approved three other national institutions to act as official quarantine agencies, especially for research material.

### **1. National Bureau of Plant Genetic Resources (NBPGR)**

The NBPGR in New Delhi and its regional station at Hyderabad in the agency involved in processing of germplasm, seed, plant material of agricultural, horticultural, and silvicultural crops of all the institutions of Indian Council of Agricultural Research (ICAR) functioning in the country. It is also responsible for quarantine clearance of seed and plant material received from International Agricultural Research Centers *viz.*, ICRISAT, ICARDA, CIMMYT, etc. ICRISAT was established in 1972 at Patancheru (near Hyderabad) to work on improvement of sorghum, pearl millet, chickpea, pigeonpea and groundnut. The quarantine clearance of all its exchanges was handled by Central Plant Protection Training Institute of Directorate of Plant Protection, Quarantine & Storage, until July 1986. This authority was later passed on to NBPGR in August 1986.

### **2. Forest Research Institute (FRI), Dehra Dun, for forestry plants and**

### **3. Botanical Survey of India (BSI) for other plants.**

Quarantine inspection, treatment and certification procedures Inspection: Inspection of plant material is an important part of plant quarantine procedure, and may be done both in the exporting country, before issue of a health certificate and after arrival to detect any pest or disease which may have become evident during transit. Publications like manuals, hand books on individual organisms of quarantine importance are prepared with illustration by each country / region to help inspectors. The following series published by Commonwealth Mycological Institute will be useful for all countries.

1. CMI descriptions of pathogenic fungi and bacteria
2. CMI/AAB descriptions of plant viruses and
3. CMI distribution maps of plant diseases.

The various steps involved in import quarantine clearance of seed and propagating plant material is outlined below

- i. Securitization of import application filed along with attached documents such as phytosanitary certificate (original), permit (importer's copy), shipping bill, invoice, packing list and customs bill of entry etc., to ensure the import is in order and that no prohibited plant material is imported.
- ii. Assessment of inspection fees and registration of application.

iii. Inspection and sampling of the consignment at port warehouses or container terminal. Sampling of seed usually carried out as per the provisions of ISTA Rules and Regulations. Whereas in case of bulk import of vegetative planting material such as cuttings/saplings/ budwoods/bulbs/tubers etc., at least a minimum of 0.1% of propagules are sampled variety and examined to ensure free from exotic pests or pathogens. In case of quarantine pests suspected, 100 per cent inspection is carried out for critical assessment of the risk.

iv. Detailed laboratory testing

a. Visual inspection: The samples of seed/ propagating plant material is examined with the help of illuminated magnifier to record live insect infestation, contamination by soil and weed seeds, nematode galls, sclerotia, smut/bunt balls etc. Sometimes inspections are carried out under U.V. lamp to facilitate detection of specific seed-borne infestation by characteristic fluorescence.

b. X-Ray test for detecting hidden insect infestation such as bruchids and weevils that bore into seed.

c. Washing test to detect surface-borne oospores of downy mildew/smut spores/ bunt spores etc. and nematode cysts. Seed samples of onion, clover and lucerne are soaked for 24 to detect stem and bulb nematode and also root washings are examined for ectoparasitic nematodes.

d. Incubation tests such as blotter test or agar plate test carried out for detecting seed-borne pathogens such as fungi. Fluorescent pseudomonas agar used for selective detection of seed-borne bacteria.

e. Grow-out test coupled with indicator inoculation tests for detecting seedborne viruses and bacteria. Besides this, special diagnostic tests such as Electron Microscopy (TEM method), Enzyme Linked Immunosorbent Assay (ELISA) are used for detection of specific viruses in the imported seed / planting material penicillinase based DAC-ELISA is widely used for the detection of virus in imported seed/plant material. The detailed testing procedures for the detection of seed-borne pathogens are outlined in the seed health testing chapter.

v. Fumigation and treatment techniques

Fumigation is the versatile technique used for eliminating insect infestation. Methyl bromide is the most commonly employed for controlling insect infestation and readily adopted in quarantine programmes as the exposure time involved is short and affect all stages of insect pests and high penetrating power. Two types of fumigation viz., i. atmospheric fumigation under gas-proof sheets or chambers and ii. vacuum fumigation in vacuum chamber is widely employed. The other

chemical treatments include insecticidal/fungicidal drippings or spraying or seed dressings are invariably associated with growing under post-entry quarantine conditions. The temperature treatments such as hot water treatment/ hot air treatment or vapour heat treatment are carried out to control internally borne infection/infestation and the latter particularly employed to control fruit fly infestation.

Cold treatments such as refrigeration to control insect infestation in fresh fruits and vegetables. Of late, irradiation is used to control insect infestation and spoilage of food products during storage and as well as application of high intensity electronic beam through an accelerator under experimentation.

### **Certification**

Phytosanitary or health certificate is a certificate which should accompany a plant or plant material or seed which is to be moved from one place to another place. This certificate indicates or certifies that the material under transit is free from pests or diseases. A model phytosanitary certificate proposed at the Government consultation on the International Plant Protection Convention at Rome in 1976 (Chock, 1977) and approved by F.A.O. in 1979 is given below.

### **MODEL PHYTOSANITARY CERTIFICATE**

(to be typed or printed in block letters)

Plant Protection Organization No. \_\_\_\_\_ of \_\_\_\_\_

To: Plant Protection Organization(s) of \_\_\_\_\_

#### **DESCRIPTION OF CONSIGNMENT**

Name and address of exporter \_\_\_\_\_

\_\_\_\_\_ Declared name and

address of consignee \_\_\_\_\_ Number

and description of packages \_\_\_\_\_ Distinguishing marks

\_\_\_\_\_ Place of origin

\_\_\_\_\_ Declared means

of conveyance \_\_\_\_\_ Declared point of entry

\_\_\_\_\_ Name of produce and

quantity declared \_\_\_\_\_ Botanical name of plants

\_\_\_\_\_



This is to certify that the plants or plant products described above have been inspected according to appropriate procedures and are considered to be free from quarantine pests and practically free from injurious pests; and that they are considered to conform to the current phytosanitary regulations of the importing country.

**DISINFESTATION AND/OR DISINFECTION TREATMENT**

Date \_\_\_\_\_ Treatment \_\_\_\_\_  
\_\_\_\_\_ Chemical (active ingredient) \_\_\_\_\_  
Duration and temperature \_\_\_\_\_ Concentration \_\_\_\_\_  
\_\_\_\_\_ Additional information \_\_\_\_\_

Additional declaration:

(Signature)

Note: No financial liability with respect to this certificate shall attach to.....(name of plant protection organization)... or to any of its officers or representatives.

**Domestic Quarantine**

Under the DIP Act, the Directorate of Plant Protection, Quarantine and storage has the responsibility to take the necessary steps and regulate the inter-state movement of plants and plant material in order to prevent the further spread of destructive insects and diseases that have already entered the country. The sole object of enforcing domestic quarantine is to prevent the spread of these diseases from infected to non-infected areas. Currently, domestic plant quarantine exists in four diseases, wart (*Synchytrium endobioticum*) of potato from 1959, bunchy top (virus) of banana from 1959, mosaic (virus) of banana from 1961 and apple scab (*Venturia inaequalis*) from 1979. Most of the states in India have plant quarantine laws to avoid entry of plant pests and diseases

**1. Bunchy top of banana:** The export and the transport from the States of Assam, Kerala, Orissa, West Bengal, Tamil Nadu to any other State of Banana plant or any other plant of the genus *Musa*, including sucker, stem, leaf, flower, and any other part thereof which may be used for propagation, or the materials of banana plant or any other plant of the genus *Musa*, which are used for packing and wrapping, excluding the banana fruit is prohibited.

**2. Banana mosaic :** The export and transport from the States of Maharashtra and Gujarat of any plant of Banana or any other plant of genus *Musa* including the sucker, stem, flower and any

other part thereof, but excluding leaf and fruit thereof is prohibited; vide Government of India notification No. F. 6-10-PPS dated the 11th April, 1961.

**3. Potato wart:** The export to potato tubers from the State of West Bengal to any other State or territory of India is prohibited.

**4. Apple scab:** The Directorate of Horticulture, Himachal Pradesh worked out a detailed scheme for the eradication of scab, and also issued a notification No. NIC.20/76 dated 28 December 1978, prohibiting the export of planting material of apple outside the State.

In Tamil Nadu as per Madras pests and Diseases Act of 1919, quarantine regulations are periodically enforced. e.g., cardamom mosaic prevalent in Anamalai area of Coimbatore District and is free from Nelliampatti area. Hence the movement of diseased plant material from Anamalai to Nelliampatti area is prevented.

### **Limitations**

There are many limitations to implementing domestic plant quarantine in India due to the vastness of the country and the unrestricted movement of plant material from one state to another. As a result the diseases like bunchy top and mosaic of banana have spread to several other states. However, the wart disease, golden nematode of potato, and scab of apple are restricted in the states where they were initially noticed.

### **Export regulations**

In India the plant quarantine measures for exporting plants and material including seeds have been streamlined and rigid inspections are enforced before the material is allowed to be landed into the country. At present plant quarantine regulations differ with different countries for major agricultural commodities that are being exported out of India. The Central Government has authorized officers of the Directorate of Plant Protection, Quarantine & Storage, ICAR Research Institutes, National Institutes like Forest Research Institute, Botanical Survey of India, and the Directorates of Agriculture of all States.

The quarantine authorities have also framed terms and conditions pertaining to inspection, fumigation or disinfection of the exportable plants and plant material in India including the following schedule/or fee for inspection and issue of phytosanitary certificate, and/or fumigation or disinfection in respect of plants, plant material, seeds, and plant products to issue phytosanitary certificate. All the plants and plant material are subjected to inspection by

officials issuing certificate. Infested materials are given necessary treatment with chemicals and fumigation if necessary.

The list of plant quarantine and fumigation stations in India is given below.

### **Punjab**

1. Plant Quarantine and Fumigation Station, Hussainiwalla, Ferozepur District.
2. Plant Quarantine and Fumigation Station, Attari–  
Wagah Border, near Attari Bus Stand, Attari, Ferozepur District.
3. Plant Quarantine and Fumigation Station, Civil Aerodrome, Rajasansi, Amritsar.

### **New Delhi**

1. Plant Quarantine and Fumigation Station, Palam Airport, New Delhi–10.
2. Plant Quarantine and Fumigation Station, Garden Reach Road, Calcutta–24.
3. Plant Quarantine and Fumigation Station Sukhiapokri, Darjeeling District.

### **Gujarat**

1. Plant Quarantine and Fumigation Station, Haryana Plot No. 75, Behind Yusuf Bagh, Bhavnagar.

### **Maharashtra**

1. Plant Quarantine and Fumigation Station, Haji Bunder Road, Sewri, Mumbai

### **Andhra Pradesh**

1. Plant Quarantine and Fumigation Station, The Harbour, Visakhapatnam–1.

### **Tamil Nadu**

1. Plant Quarantine and Fumigation Station, 6, Clive Battery, Chennai–1.
2. Plant Quarantine and Fumigation Station, 335, Beach Road, Tuticorin –1.
3. Plant Quarantine and Fumigation Station, Tiruchirappalli Airport, Tiruchirappalli.
4. Plant Quarantine and Fumigation Station, 110, Railway Feeder Road, Rameswaram.

### **Kerala**

1. Plant Quarantine and Fumigation Station, Willingdon Island, Cochin– 3

**Cultural methods – Rouging, eradication of alternate and collateral hosts, crop rotation, manure and fertilizer management, mixed cropping, sanitation, hot weather ploughing, soil amendments, time of sowing, seed rate and plant density, irrigation and drainage**

### **Eradication**

Eradication is the elimination of pathogen after it has become established in the area where host is growing. The following are the important methods followed to prevent the spread of the disease:

- i. eradication of alternate hosts,
- ii. eradication of collateral and self-sown overwintering hosts
- iii. eradication of the affected plants or trees,
- iv. eradication of pathogens from infected plant parts by surgery and
- v. eradication of culled out plant materials, debris, etc., through different cultural practices

#### **i. Eradication of alternate hosts**

Removal of alternate hosts helps to prevent and check the spread of the disease caused by heteroecious rust pathogens in the primary hosts. Barberry bush is the alternate host for black stem rust pathogen *Puccinia graminis tritici* on wheat where the pathogen survives in the off-season. Barberry was eradicated in Canada, Denmark, France, Hungary, Norway and in the U.S.A. by passing stringent laws in each country. The eradication of barberry had two benefits i.e., it elimination of early spring primary inoculum and prevention of the formation of new physiologic races of the pathogens. In the U.S.A. white pine blister rust (*Cronartium ribicola*) was controlled by eradication of alternate host, *Ribes*. In Australia, Europe and the U.S.A. the apple rust (*Gymnosporangium juniperi-virginianae*) is controlled by eradication of alternate host, cedar.

#### **ii. Eradication of collateral and self-sown overwintering hosts**

There are many weed hosts or wild species of cultivated plants act as collateral hosts or volunteer plants of an economic crop which act as reservoirs of pathogens of annual crop. Reservoir hosts help the pathogen to continue the infection chain. The primary inoculum is produced on and dispersed from these hosts to the cultivated crop hosts. If these wild or uneconomic host plants of the pathogen are destroyed, the sources of primary inoculum are

eliminated and chances of initiation of the disease in the crop hosts are reduced. Destruction of these hosts breaks the life cycle of the pathogen and the infection chain. Reservoir hosts are indigenous plant species which are not actually involved with the life cycle of the pathogen but provide additional sites for its persistence and multiplication. In some cases such plant species act as symptomless carriers, especially for viruses and root pathogens. Regional elimination of such hosts requires careful attention to roadside areas and other non-agricultural land also.

<b>Crop</b>	<b>Disease</b>	<b>Pathogen</b>	<b>Collateral hosts</b>
<b>a. Fungi</b> 1. Rice	<b>Blast</b>	<i>Pyricularia oryzae</i>	<i>Brachiaria mutica</i> , <i>Dinebra retr oflexa</i> , <i>Leersia hexandra</i> , <i>Panicum repens</i> .
<b>2. Sorghum</b>	Ergot	<i>Sphacelia sorghi</i>	<i>Panicum</i> spp.
<b>b. Bacteria</b> 1. Rice	Bacterial leaf blight	<i>Xanthomonas oryzae</i> pv. <i>oryzae</i>	<i>Cyanodon dactylon</i> , <i>Cyperus rotundus</i> , <i>Leersia hexandra</i> , <i>Leersia oryzoides</i> , <i>Panicum repens</i> , <i>Paspalum dictum</i> .
2. Apple and pear	Fire blight	<i>Erwinia amylovora</i>	<i>Hawthorn</i> bushes <i>Crataegus</i> sp.
3. Cotton	Bacterial blight	<i>X. axonopodis</i> pv. <i>malvacearum</i>	<i>Eriodendron anfructuosum</i> , <i>Jatropha curcas</i> , <i>Thurberia thespoides</i>
<b>c. Viruses</b> 1. Potato	Rugose mosaic	Rugose mosaic virus	<i>Physalis</i> spp.

2.Bean	Yellow mosaic	Beanyellow mosaicvirus	Sweetclover
3.Bhendi	Yellow veinmosaic	Bhendiyellow vein mosaic virus	<i>Hibiscustetraphyllus</i>
<b>d.Phytoplasma</b> <b>1. Brinjal</b>	Littleleaf	Phytoplasma	<i>Catharanthusroseus, Datura sp.</i>

Self sown crops / volunteer plants help the pathogen to overwinter / oversummer in the absence of economic hosts. In Sudan it was enforced through legislation to pull out the cotton plants to prevent regrowth which facilitate the carryover of the cotton leaf curl virus. Wheat streak mosaic virus has been effectively controlled by eliminating the volunteer wheat plants that served as reservoirs for the virus.

**iii. Eradication of affected plants or trees**

In some threatening plant diseases, it is essential to eradicate the host and the pathogen from an area. Citrus canker (*Xanthomonas axonopodis* pv. *citri*) is an example of success of an eradication programme. This disease was first noticed in Florida citrus trees in 1913. An eradication campaign was started in 1915. All the citrus nurseries and orchards were inspected and the infected trees were cut and burnt. The eradication programme continued till 1927 and no citrus canker was present in that area. Peach yellows and peach rosette were also controlled by removal and destruction of diseased trees. In Tamil Nadu also there were some eradication campaigns launched under Destructive Pests and Diseases Act. Eradication programme was setup to control bud rot of palms and completed with success. At Sathyamangalam eradication of sandalwood tree affected by spike disease was also made to contain this disease.

**iv. Eradication of pathogens from infected plant parts by surgery**

Eradication of affected plant parts (tree surgery) are also practiced in certain cases which reduces the source of primary inoculum. Lesions caused by fire blight bacterium (*Erwinia amylovora*) on pear and apple trees are removed during winter months. This not only prevents further spread in the affected trees but also reduces the amount of inoculum that can spread to other branches and trees. The cankered areas in the branch or trunk of almond and

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pear trees caused by *Ceratocystis fimbriata* are surgically removed and the trees are saved. Tree surgery is

also practiced in coconut trees affected by stem bleeding disease (*Ceratocystis paradoxa*), citrus gummosis (*Phytophthora citrophthora*), *Dendrophthoe* spp. on citrus, bud rot of palms (*Phytophthora palmivora*) and kolerog of arecanut (*P. arecae*)

**v. Eradication of culled out plant materials, debris etc. through different cultural practices**

**2. Croprootation**

Croprorotation is essentially a preventive measure and has its effect mainly on the succeeding crop. Crop rotation is the oldest and cheapest method adopted in agriculture for eradication of certain types of pathogens from infested soil. Continuous cropping or monoculturing provides opportunity for perpetuation of pathogenic organisms in the soil when the same crop is raised year after year in the same field.

The soil-borne pathogens of that crop easily perennate in the soil and increase in their population. After some time, the soil becomes so heavily infested that it becomes unfit for cultivation of the particular crop. Virus diseases of crop plants and their vectors are found to increase after every crop if a crop is cultivated continuously in a field. On the other hand, when immune, resistant or non-host crops are grown for a definite duration after a susceptible crop in the field it is expected that in the absence of nutrition, the pathogen will be starved off and the population of such pathogens consequently decreases.

It is also possible that different crops release some biochemical substances in their root exudates which either directly kill the pathogen or encourage development of antagonistic microorganisms in the soil. In this way, crop rotation is one of the most effective methods of root disease control. Organisms which are soil inhabitants remain in soil for a very long time, even more than five years in the absence of the host. Long-lived spores or the organisms by themselves, subsist as saprophytes and therefore their presence in soil is long term. Onion smut (*Urocystis cepulae*) and club root (*Plasmodiophora brassicae*) organisms are producing resistant type of spores while *Rhizoctonia*, *Fusarium* and some species of *Pythium* are those which could remain in soil as saprophytes for a very long time.

Eradication of such organisms becomes fairly difficult. Soil also harbours soil invaders. These organisms are not persistent and they can live as long as the host residues serve as substrate. They perish when they are forced to exist in the soil in competition with soil inhabitants and disappear gradually in due course. Bean anthracnose fungus *Colletotrichum lindemuthianum*, cabbage black rot bacterium, *Xanthomonas campestris* sp. *campestris* are



some examples, which live in soil for 1 to 2 years. They can be eliminated from soil by adopting 3 or 4 year rotation with non-host crops. Crop rotation is effective in the control of brown stem rot of soybean (*Cephalosporium gregatum*). The disease incidence can be reduced to a great extent by rotating with corn for 4 to 5 years between two soybean crops.

Crop rotation with sugarcane or paddy is effective in the control of ‘Panama wilt’ of banana (*Fusarium oxysporum* f.sp. *cubense*) and crop rotation with paddy or green manure is effective in the control of red rot of sugarcane (*Colletotrichum falcatum*). Rotation of cereal crops like pearl millet, finger millet or fox-

tail millet is recommended for the control of *Macrophomina* root rot of pulse crops. Two year crop rotation with lucerne is recommended in the control of *Verticillium* wilt of cotton. Many diseases such as *Fusarium* wilt of pigeon pea (*F. udum*), foot rot of betel vine (*Phytophthora capsici*), bacterial leaf blight of rice (*Xanthomonas oryzae* pv. *oryzae*), bacterial blight of cotton (*X. campestris* pv. *malvacearum*) etc., are controlled by this method. Soybean seed infection by *Phomopsis* sp. can be reduced by rotating soybean with maize. Pathogens are reduced or eliminated by following the crop rotations given in the table.

**Table. Effect of crop rotation in reduction/elimination of plant pathogens**

Beneficial crop	Pathogen reduced or eliminated	Preceding crop
1. Rice	<i>Verticillium dahliae</i>	
	Cotton	2. Pea
	<i>Gaeiimannomyces graminis</i>	
	Wheat	3. Sudangrass
	<i>Pseudomonas solanacearum</i>	Tomato

### 3. Fallowing

Fallowing starves the pathogen and helps in reduction of the inoculum by elimination of the host. Diseases like *Macrophomina* root rot on different crop plants is controlled by following this method. Flood fallowing is to a depth of 0.6 to 1.5 m for 4 to 6 months markedly reduced the Panama wilt pathogen *Fusarium oxysporum* f.sp. *cubense* inoculum in banana. Soil inoculum of *Phytophthora parasitica* var. *nicotianae*, the causal organism of black shank of tobacco was destroyed by flooding the field for 3 to 4 months and by raising swamp rice in a 2 year rotation with tobacco-rice crop in Java. Flooding the soil strewn with debris infected by *Xanthomonas*

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*axonopodis* sp. *malvacearum* for 4 days reduced the inoculum level and thus the incidence of disease was only 2.1% as against 69.5% in unflooded fields. Wet fallowing makes

the pathogenic propagule in or on the soil to germinate, spent them, is become susceptible attack of saprophytes. Example, *Sclerotium rolfsii* and *Verticillium dahliae*. The sclerotia or microsclerotia of these fungi are activated in the absence of root exudates of this host. They germinate quickly when there is alternate wetting and drying of the soil. When the population of *Pythium myriotylum* is not high wet fallowing is successful in reducing the population. Wet fallowing reduces saprophytic survival of *Alternaria solani* on crop debris.

#### **4. Application of organic manures**

Addition of organic manures like farm yard manure or green manures or oil cakes to the soil increases the antagonistic microorganisms in the soil. Build up of antagonistic microorganisms reduces the population of soil borne plant pathogens and the diseases caused by them. Application of farm yard manure at the rate of 12.5 tonnes/ha reduced the incidence of *Macrophomina* root rot of cotton. Application of 5 kg of neem cake/tree three times in a year reduces the basal stem rot (*Ganoderma lucidum*) of coconut. In the control of sesame root rot (*Macrophomina phaseolina*) application of neem cake at the rate of 150 kg/ha is recommended. Application of neem cake at the rate of 2 tonnes/ha in two split doses and covering with mud reduced foot rot disease in betel vine garden.

#### **Soil amendment**

It has been proved that the organic amendments rich in carbon and deficient in nitrogen control the take-all disease (*Ophiobolus graminis*) of wheat. There is considerable liberation of CO<sub>2</sub> by soil saprophytes which suppresses the pathogenic activity of this fungus. In the process of survival also, low nitrogen content in the soil reduces the longevity of the fungus. *Phytophthora* root rot of avocado is controlled by amending the soil with alfalfa meal—a material of low C/N ratio. The other diseases are pea root rot *Aphanomyces euteichus* when cruciferous plant residues were incorporated into the soil. Alfalfa meal and barley straw application in the soil reduced the root rot of cotton and sorghum caused by *Macrophomina phaseolina*. Black scurf of potato (*Rhizoctonia solani*) is less in the field where wheat straw was incorporated.

#### **5. Summer ploughing**

Deep ploughing during summer periods buries the inocula of fungi of soil borne nature. Fungal propagules, sclerotia and different types of spores conidia on plant refuse die when exposed to sunlight due to the high temperature prevailing during the summer. Further

infected self sown plants, volunteer hosts plants, weed hosts, regrowths from the plant roots, alternate hosts and alternative hosts are also destroyed. Here, the spread of the disease is avoided. Groundnut blight (*Corticium rolfsii*) is controlled by ploughing the soil to a depth of 20cm. The inverted plough sole soil buries the sclerotia of the fungi, *Claviceps*, *Sclerotium* and *Sclerotinia* in association with plant or alone, impedes the discharge of ascospores from perithecia. Bunt and smut spores of wheat, smut spores of sugarcane and sorghum and microsclerotia of *Verticillium* in cotton are buried deep in the soil by deep ploughing.

## **6. Crop growing seasons**

Rice blast becomes serious when the rice crop is raised from August to September in Tamil Nadu. Ragi blast becomes serious when sowing is made between June and August. Similarly yellow mosaic of blackgram/green gram and phyllody of sesame are serious during kharif season in South India. Incidence of powdery mildews of different crops is found to be high during rabi when compared to kharif and summer seasons. In bhendi, yellow vein mosaic incidence is very high during summer. The seasons with high incidence of diseases should be avoided in the epidemic areas.

### **a. Adjustment of sowing time**

In many diseases the incidence is more severe when the susceptible stage of the plant growth and favourable conditions for the pathogen coincide. While choosing the time of sowing it should be taken into consideration that susceptible stage of the crop growth and soil conditions and other environments favourable for maximum activity of the pathogen does not fall at the same time. Properly adjusting the sowing dates can give good dividends. Late planted wheat crops contract less infection than wheat planted on normal dates. Early and late sown crops have been found to be free from Oodhubathi disease of rice.

Avoiding cool and cloudy days for planting will help to reduce red rot of sugarcane. Late sowing of winter wheat and barley is considered to be the most effective measures in reducing take all disease of wheat. Rapeseed sown in mid to late August is more liable to attack by leafspot (*Alternaria brassicae*) than late-sown crops. Pea and gram planted soon after rains when soil temperature and moisture are at a high level, show high incidence of root rot and blight. As the soil temperature falls and moisture becomes less (Nov-Dec) these diseases are also reduced. In areas where these diseases are serious, late sowing helps in saving the crop. Stem rust of wheat damages the late sown crop more than the early sown crop. Because, time of onset of

disease and ear formation coincides. Sowing from January to April or October to December is advocated to escape from the attack of neck blast of finger millet. Peas and chickpea sown in October usually suffer heavily from root rot and wilt (a complex of *Fusarium*, *Rhizoctonia* and *Sclerotium*). When these crops are sown late, the diseases are not so severe or almost absent. The groundnut rosette is transmitted by *Aphis craccivora*.

In Nigeria the population of this vector is low in crops sown in June than in July. The sowing time is adjusted inumbu and sorghum in such a way that the flowering stage does not coincide with the rainy season to avoid the sugary diseases. Early sown crops show decreased incidence of curly top and yellows on sugarbeet, rosette on groundnut and barley yellow dwarf on cereals. Delayed sowing on the other hand is beneficial to maize rough dwarf disease.

### **b. Adjustment of harvesting time**

Harvesting of groundnut should not coincide with the rainy days and it helps to avoid infection by *Aspergillus flavus*. Freedom of onions and roses grown in rainless seasons from downy mildew diseases and freedom of beans, chilli and cucurbits from bacterial diseases in such seasons are the best examples for sowing of crops at correct season to avoid disease outbreaks. In the case of deciduous fruit trees and grapevines, the season of sprouting, flowering and fruit set can be advanced or delayed by pruning practices or by treatments to break dormancy. Advantage can sometimes be taken of this fact to avoid coincidence of all or one of these phases of host growth with weather periods particularly favourable to specific pathogens that attack trees in the phases.

## **7. Growing of seed crops**

Coffee can be grown in the western Hemisphere usually free from coffee rust which causes heavy losses in Eastern Hemisphere. In the case of virus diseases this will be more useful. By growing seed materials in isolated places where the population of vectors is very low and the condition is uncongenial for the vectors. Virus free potato tubers to be used as seeds are grown in cool and windy places in many parts of the world. Under tropical and subtropical countries, such conditions prevail in the hills at high altitudes. Obtaining seed from disease-free localities has been very successfully resorted to the elimination of many seed-borne diseases. In the U.S.A. seed-potatoes are invariably grown in northern snow-clad sections, where viruses are practically absent and then exported to various other sectors in the south. Similar practice has been in vogue in India, where seed-potatoes are annually imported in southern states from Simla

hills for control of virus diseases and bacterial ring. In the U.S.A, the seed growing areas have been shifted to arid pacific regions for crops like cabbage, turnip, beans and peas for obtaining disease-free seed and indirectly controlling such diseases like black leg and black rot of cabbage and turnip and anthracnose of beans and peas. Similar practice is obtained in parts of Bombay, where the foot rot of ginger (*Pythium myriotylum*) prevalent in the southern parts, is controlled through the importation of seed-rhizomes from disease-free arid regions of the north, where the disease is practically non-existent on account of the dry climate, lighter soils and moderate rainfall.

### **8. Selection of seeds and seed materials**

Seeds and seed materials carry many fungi, bacteria, viruses and phytoplasmas and may introduce these pathogens into the field, i.e., seeds and seed materials form the primary source of infection. Seed and seed materials like cuttings, tubers, grafts, setts etc., should be well matured, disease free, uninjured and have a high germinating capacity. The absence of an initial inoculum in seeds is definitely helpful in delaying or suppressing the incidence of the disease. It is a preventive method.

The diseases like foot rot, brown spot, short smut of sorghum, loose smut of wheat, bacterial blight of rice, bacterial blight of cotton, leaf crinkle of blackgram etc., are transmitted through seeds. Virus diseases and black ring of potatoes, foot rot of ginger, foot rot of betelvine, Panama disease of banana, red rot of sugarcane cassava mosaic, bunchy top and virus diseases of fruit trees are transmitted through tubers, setts, rhizomes, corns, grafts and budwoods. 'Tuber indexing' is a special method to obtain disease free seed materials in potato. It is commonly practiced by nurseries and seed merchants selling potato seed tubers. Use of seeds in the place of rhizome/sucker is recommended in the control of 'katte' disease of cardamom.

### **9. Leveling of the field and provision of drainage facilities**

Water stagnation in different patches of field favours the fungi like *Pythium*, *Phytophthora*, *Rhizoctonia solani*, etc., for which proper leveling of the field before sowing or planting is very essential. Further improving the drainage is necessary in the control of sheath blight of rice. Provision of drainage channels in orchard crops like citrus, jack, mango etc., in the garden is necessary before planting. In the control of damping-off diseases of vegetable and other crops, raising seedling in the raised beds method is followed. Foot rot of ginger (*Pythium myriotylum*) is also controlled by following the raised bed system of nursery.

### **10. Seed rate**

Use of higher seed rate in the nursery creates favourable microclimate for the pathogens causing damping-off in vegetables, tobacco, chillies and forest nurseries. Hence, use of optimum seed rates should be adhered in such crops.

### **11. Burning of stubbles and crop residues**

Burning of plant wastes, crop residues, stubbles, *etc.*, in the areas selected for raising nurseries for vegetable crops, tobacco, chillies and forest trees *etc.* heats the soil and kills the inoculum of the pathogens present in the top layer of the soil. When nurseries are raised in these areas incidence of damping off disease is highly reduced. This practice is also followed in pits made for planting coconut, banana, fruit trees *etc.*, Burning of wheat plant every second or third year is suggested for eradication of pathogen in the field when *Cephalosporium gramineum* infects wheat. Otherwise, debris in the field helps the perpetuation of the pathogen and the disease. Burning of rice crop residues avoid carryover of sheath blight (*Rhizoctonia solani*); stem rot (*Sclerotium oryzae*) of rice and bacterial blight of cotton.

### **12. Depth of sowing**

Depth of sowing greatly influences seed transmission of smuts. Shallow planting in wet soils protects wheat plants from *Urocystis tritici* (flag smut) of wheat. Deep planting may cause delay in the emergence of seedlings, which may be vulnerable to pre-emergence damping off. Early emergence results in early lignification of tissues which become resistant to attack of soil-borne pathogens.

### **13. Spacing**

Closer spacing invariably alters the microclimate underneath the canopy of the crop which may provide favourable environment for development of diseases. Boll rot in cotton is quite common in crowded crop. Defoliation of plants or skip cropping gives better control against the boll rot disease. In certain virus diseases like groundnut rosette the incidence is observed to be less when wider spacing is adopted. Closer spacing favours many air borne diseases because of high humidity in the crop canopy. Early and late blight of groundnut and blister blight of tea are more in dense canopy. Early spread of black rot of cabbage takes place in closer spacing. Crowded stands may reduce some systemic diseases. Cotton wilt caused by *Verticillium albo-atrum* will be less in closely planted crop if the fungal inoculum is less in the soil. Similarly closer spacing of rice reduces rice tungro virus infection particularly when vector

population is less. Avoiding shade and providing wider spacing reduce the incidence of powdery mildew of tobacco. Late blight of potato and downy mildew of grapevine spread fast in closer spaced crops. In the case of bud necrosis of groundnut caused by tomato spotted wilt virus, seeds are sown adopting closer spacing of 15x15cm to compensate the rogued out plants with regard to plant population and yield. These are examples where dense sowing helps in disease reduction.

The virus of tomato leaf curl, transmitted by *Bemisia tabaci*, is less severe in a crowded planting than in spaced planting. Same is true for cucumber mosaic, transmitted by *Aphis gossypii* and groundnut rosette transmitted by *Aphis craccivora*. The fungal diseases for which the phenomenon of flower incidence at closer spacing of the crop has been studied most profitably is the wilt caused by *Verticillium albo atrum* and *V. dahliae* in cotton. This is ascribed to the reduction of effective inoculum per plant in proportion to the increase in the number of plants per unit area in the densely sown field. The incidence of brown rot (*Cephalosporium gragatum*) of soybean is also higher in widely spaced planting than in closer rows.

#### **14. Method of sowing/planting**

In places where water accumulation is a problem to the crop growth sowing of seeds on the sides or ridges is found effective in reducing the incidence of *Sclerotium rolfsii* on groundnut and vegetable crops and *Sclerotinia sclerotiorum* and *Rhizoctonia solani* on vegetable crops and *Phytophthora* blight of pigeon pea. High ridging prevents infection of potato tubers, by zoospores from leaf lesions in late blight diseases. Ridging is disadvantageous in water deficit areas where it encourages pathogens like *Macrophomina phaseolina*.

#### **15. High budding**

High budding is a practice to avoid infection by gummosis fungus of citrus trees. In low budded plants the bud point is close proximity to infection centre (the soil), become readily diseased. High budding is a simple device for lengthening this distance between the bud point and infected soil. In this method the soil borne pathogens (*Phytophthora palmivora* and *P. citrophthora*) have no chance of reaching the bud point, through which they enter the bark. Staking of lower most branches arising close to the soil, increases the distance between the fruits and soil inoculum and removes the chances of brown rot (*Phytophthora sp*) infection and buck-eye rot of tomato (*P. nicotianae* var. *parasitica*).



## **16. Avoiding injury**

Injury of plant parts should be avoided in order to check the entry of pathogens. Clipping of tips of tall rice seedlings favours the entry of bacterial blight pathogen and incidence of the disease. Hence clipping should be avoided at the time of transplanting of rice. While harvesting the pods in groundnut, fruits in tree crops and vegetable crops injuries to the fruits pave the way for the pathogen and causing pod/fruit rot. It also reduces the storage life of fruits and vegetables. Hence much care should be given to avoid wounds during the harvest time.

## **17. Altering the soil pH**

In certain soil-borne diseases adjustment of soil reaction helps in the reduction of inoculum level of pathogens. The altered pH of the environment forms a barrier against the pathogen. A very low pH less than 5.2 is unfavourable to common scab bacterium on potato (*Streptomyces scabies*). Thus, use of acid-forming fertilizers (like sulphur) and avoiding lime and calcium ammonium nitrate application are effective in controlling the common scab disease.

On the other hand the club root pathogen of cabbage (*Plasmodiophora brassicae*) cannot live and infect when the soil pH is 7.0 or more. Hence liming which increases the soil pH gives satisfactory control of club root disease. In Punjab, root rot of tobacco (*Macrophomina phaseolina*) has been overcome by application of 2.5 to 5.0 tons of lime/ha to the soil.

## **18. Mixed cropping**

Mixed cropping materially helps in checking certain diseases. Blight of pulse crop (*Phyllosticta phaseolina*) has been successfully overcome by growing pulses as a mixed crop with cereals like sorghum and pearl millet.

## **19. Intercropping**

Intercropping is also a device in the control of some soil-borne diseases. Intercrops should be properly chosen so that they should not have any common pathogen. For example, *Macrophomina phaseolina* has got wide host range and hence common host should not be grown as intercrops. Intercropping with moth bean (*Phaseolus aconitifolius*) in a cotton field reduced the root rot (*M. phaseolina*) incidence.

Due to reduction in the number of host plants there is sufficient spacing between them and chances of contact between foliage of roots of diseased and healthy plants are greatly reduced. Therefore, root pathogens are unable to spread from diseased to healthy roots and spread of foliar pathogens is also reduced to a great extent. Intercropping of sorghum in

pigeonpea field reduced the wilt (*F. udum*) incidence. The roots of non-host plants may act as a barrier obstructing the movement of pathogens in soil. They may release toxic substances from their roots which may suppress the growth of the pathogens attacking the main crop. Hydrocyanic acid (HCN) in root exudates of sorghum is toxic to *F. udum*, the pigeonpea wilt fungus. Intercropping of sorghum or mothbean in a crop of clusterbean reduced the incidence of root rot (*R. solani*) and wilt (*F. coeruleum*) from 50 to 60% in single crop to 8 to 15% in the mixed crop.

Intercropping of pigeonpea with gingelly at 1:6 ratio reduced the incidence of phyllody disease. In Jordan, intercropping tomatoes with cucumber is found to be effective and cheaper in controlling the whiteflies and lowering the incidence of tomato yellow leaf curl virus (TYLCV). Cucumber is planted one month before tomato. Cucumber is known to be a preferred host for whiteflies and immune to TYLCV. Insecticides are applied when adult whitefly populations are at high levels, usually two weeks after planting of cucumber and the second one before tomato planting. Growing of an intercrop of cereals such as corn or sorghum between rows of peach trees is an effective method in combating Texas root rot (*Phymatotrichum omnivorum*) infection in the U.S.A.

## **20. Barrier cropping**

Taller crops can be grown to protect a crop of lesser height from virus vectors. The insects may land at the taller crops (barrier crops) and the dwarf crop may escape from virus diseases by those insects. Barrier cropping with 3 rows of maize or sorghum or pearl millet around the main crop namely black gram or green gram is effective in reducing the vector population and incidence of yellow mosaic. Another best example is growing of 3 rows of kale or barley as barrier crops in cauliflower seedbeds and undersown beetsteckling against cauliflower mosaic and beet yellows diseases respectively. The incoming aphids are thought to land on the barley or kale and probe briefly, causing them to lose the non-persistently transmitted virus they are carrying. Maize or sunflower are the other barrier crops considered for these crops.

## **21. Decoy crop and trap crop**

Decoy crops (hostile crops) are non-host crops sown with the purpose of making soil-borne pathogens waste their infection potential. This is effected by activating dormant

propagules

offungi, seedsof parasitic plants, etc. in absence of the host. A list of pathogen that can be decoyed is given in table.

**Table. Decoy crops for the reduction of pathogen populations**

<b>Host</b>	<b>Pathogen</b>	<b>Decoy crops</b>
1. Sorghum	<i>Striga asiatica</i>	Sudangrass
2. Cabbage	<i>Plasmodiophora brassicae</i>	Ryegrass, <i>Papaver rhoeas</i> , <i>Reseda odorata</i>
3. Potato	<i>Spongopora subterranea</i>	<i>Datura stramonium</i>
4. Tomato, tobacco	<i>Orobancha</i> spp.	Sunflower, safflower, lucerne, chickpea etc.

Trap crops are host crops of the pathogen, sown to attract pathogens but destined to be harvested or destroyed before they complete their life cycle. Fodder sorghum can be raised as a trap crop to reduce down mildew of sorghum.

## **22. Trenching**

Trenching between rows of trees in orchards has been effectively utilized in arresting the growth and spread of the pathogen in the soil to the neighbouring trees. *Ganoderma lucidum* root rot infected citrus trees should be isolated by digging a trench of 30 cm wide and 60 cm to 90 cm deep around the tree at a distance of 2.5 to 3.0 m from the base to prevent the contact of diseased roots with healthy roots. Thereby the spread of the pathogen to neighbouring tree is prevented. Similar method is also followed in the control of basal stem rot (*Ganoderma lucidum*) of coconut in India.

## **23. Isolation distances**

The distance between seed production and commercial plots has been worked out for reducing seed borne loose smut of barley and wheat. Barley and wheat crops should be isolated by at least 50 m from any source of loose smut infection for production of certified seeds in the U.K.

The number of viruliferous insects reaching a healthy crop from a diseased one decreases with distance between them so that cultivation of susceptible crops at a distance from each other delays or reduces the severity of virus diseases. Incidence of lettuce and cucumber mosaic virus is about 3% if the new lettuce crop is sown 0.8 km away from an old lettuce field. Much

greater incidence of mosaic in sugarbeet fields occurs within 90 metres of a seed crop than in the fields at a greater distance. Beet mosaic and beet yellows are markedly reduced by isolating beet fields by 19 to 24 km and 24 to 32 km respectively from a large source of infected beets.

#### **24. Yellow sticky traps**

Sticky, yellow polythene sheets erected vertically on the windward side of red pepper fields have been shown to reduce the incidence of potato virus Y (PVY) and cucumber mosaic virus (CMV) in the crop. The aphids are attracted to the yellow colour and are caught on the sticky polythene. The control obtained was so successful that the method has become a standard control procedure in red pepper crops in Israel. Similar traps have also been used to protect 'seed' potato crops, against potato leaf roll virus. Yellow sticky traps are in use to attract and kill the whitefly vectors which spread yellow mosaic of blackgram and greengram and bhendi yellow vein mosaic.

#### **25. Mulching**

Mulching or covering of top soil with organic residues often helps in reducing plant diseases. Mulches of non-host origin should be used in the field. These mulches are known to release inhibitory substances in the underlying soil and also promote development of parasites and predators of nematodes. Reflective surfaces (mulches) laid on the soil around the crop plant, have been found to be highly effective in controlling aphid vectors. Aluminium strips or grey or white plastic sheets are used as mulch and it has successfully protected red peppers against CMV and PVY in Israel and summer squash against watermelon mosaic virus in the Imperial valley of California. Straw mulches have been successfully used to control the white fly-transmitted tomato yellow leaf curl virus in tomato crops in Israel. It is believed that the colour of the straw attracts the whiteflies and they are subsequently killed by the reflective heat. The disadvantage with straw mulches is that they eventually lose their yellow colour, but prolonged control may be obtained if straw is replaced by yellow polythene sheets.

#### **26. Irrigation water management**

Irrigation to the crop in the field is to wet the soil to the extent that roots easily get water and nutrients. If excess water is added to soil, it may directly affect activity of pathogens and/or it may affect disease incidence through the effect on the host. Scab attack on potato tubers is prevented by maintaining soil moisture near field capacity during tuber formation. Bacterial flora antagonistic to *Streptomyces scabies* increases under high moisture conditions. The

charcoal rot pathogen, *Macrophomina phaseolina* attacks potatoes and cotton when the soil temperature rises and there is water stress. By irrigating the field, soil temperature is brought down, stress is removed and the disease is suppressed. When excess irrigation is made the juvenile stage of plants is lengthened making it susceptible to attack of fungi like *Pythium*. Supply of frequent but low quantity of irrigation water is, therefore, recommended for reducing chances of damping off in nurseries.

Under conditions of excess water, respiration of roots is inhibited and many soluble salts accumulate in toxic amounts around the roots and base of the stem. This increases disease proneness of the roots. Irrigation increases guttation. Guttation drops on leaves serve as media for multiplication and penetration of many pathogens, such as *Helminthosporium* spp. on cereals and *Xanthomonas campestris* on *Brassica* spp. Cereal rusts usually are more severe when the crop is grown in wet soil than in relatively drier soils. Vascular wilts appear aggravated soon after irrigation. These effects are through the host.

Pathogens directly taking advantage of excess water are those that need wet soil for (i) activation of their resting structures and (ii) for movement of these propagules. Thus, in presence of excess free water bacterial cells and zoospores of Pythiaceae fungi are dispersed easily. Therefore, at the plant stage when these pathogens can attack the crop irrigation should be avoided. Generally, sprinkler irrigation increases diseases by increasing leaf wetness and by dispersing propagules of the pathogens by water splashes just like rain water. At the same time, it has some advantages also such as washing off inoculum from the leaf surface.

Irrigation especially at seed-development stage, may favour seed infection. Irrigation time and amount of water should be controlled so that the relative humidity is not raised to such an extent that it becomes conducive for seed infection. Control of seed-borne diseases favoured by wet climate can be achieved by raising the seed crop in dry areas. Some examples are anthracnose of bean and cucurbits (*Colletotrichum* spp.), Ascochyta blight of pea (*Ascochyta* spp.) and bacterial blight of legumes. Such crops can be grown in dry areas with the help of irrigation so that these aerial parts remain dry and do not contact infection. Virus-free potato seed tubers can be produced more successfully in areas where temperature and moisture conditions do not favour buildup of populations of the insect vectors.

Sclerotia, smut spores, chlamydospores, oospores and mycelium found in the soil are carried from one field to another through irrigation and drainage water. Stem rot, sheath blight and bacterial blight diseases of rice, damping off of vegetables and *Macrophomina* root rots of many crops spread mainly through irrigation and drainage water. Hence care should be taken not to irrigate a healthy field using drainage/irrigation water from a diseased field.

## **27. Field and plant sanitation**

Field and plant sanitation is an important method of disease control through cultural practices. The inoculum present on field plants in the field may multiply on the plant or in the soil and in due course of time may be sufficient to nullify or reduce the effect of control practices. Many pathogens overwinter or oversummer on plant debris during the off-seasons and become active when the crop is again grown in the field. Hence plants bearing pathogens or plant debris introducing inoculum into the soil should be removed as early as possible. In most of the soil borne diseases like wilt and root rot, it has been reported that as long as the dead roots and other roots and other affected parts are present in the soil, the fungus continues its growth. When such diseased plant materials are removed, there is a quick decline in the population of pathogens in the soil.

In this manner *Fusarium* wilt of cotton, pigeon pea and banana, *Verticillium* wilt of cotton, root rot of beans, downy mildew of pearl millet, sorghum, maize and peas, foot rot of betel vine, bacterial blight of cotton, white rust of crucifers, black spot of rose, powdery mildew of pea and cereals are reduced. In certain areas the linseed rust fungus (*Melampsora lini*), the rice blast and brown spot fungi and the fungus causing early blight of potato also persist through dormant stages in diseased crop debris. Destruction of crop debris by burning immediately after harvest reduces the amount of inocula which survive through debris.

It has been observed that leaf blight disease of rice particularly one caused by *Helminthosporium oryzae* is carried over in the stubbles and primary infection is evident in the self-sown tillers arising from these stubbles. Infection of *Sclerotium rolfsii* on jute is carried over in the foot and root regions in the stubbles left over after harvest of the jute plants. Sugar cane stubbles left over in the field help to carry over red rot fungus *Glomerella tucumanensis*, *Xanthomonas oryzae* pv. *oryzae* causing bacterial leaf blight disease on rice is capable of surviving for some time in rice stubbles. In many cases, diseased planting materials left in the field after discarding them, serve as sources of infection as in the case of late blight of potato

where piles or refuse of rejected tubers later become an important source of infection. Left over plant parts of maize infected with the smut *Ustilago zae* constitute an important source of infection later.

Avoidance of the transfer of inoculum from one field to another by man, machine or water is one of the ground rules of cultural control. Where soil-borne diseases are concerned, anything that carries soil is suspect, this includes wheels, boots and water flowing either from adjacent fields, or through drainage ditches from distant fields. As regards sap-borne viruses, attention must be paid to disinfection of wheels and of the hands of labourers, as they pass from one field to another. Where such virus can also be carried on clothing. The work should be planned so that the labourers do not go from old to younger fields on the same day.

Many pathogens are capable of surviving on implements and materials used in sequential seasons. Tobacco mosaic virus has been shown to survive on iron stakes used for tomato trellises and disinfection of such stakes has been recommended. Soil adhering to plastic sheeting may carry sclerotia and other overwintering bodies.

## **28. Roguing**

Roguing consists of completely removing or uprooting the diseased plants to prevent further spread of the disease. This method is widely adopted in the control of virus diseases spread by insects (cassava mosaic, yellow mosaic of blackgram and green gram, citrus tristeza, katte disease of cardamom, bunchy top of banana) and basal stem rot of coconut, green ear

of pearl millet and broomrape (*Orobanche*) in tobacco. The whipsmut of sugarcane (*Ustilago scitaminea*) in the canal areas of Bombay in Co.475 variety has been greatly checked by roguing carried out over wide areas and long period. In Jamaica, a country-wide campaign of destroying infected plants has succeeded in the control of Panama wilt of banana. Root rot and wilt attacked plants after their death should be as and when noticed in the field uprooted and burnt to check the inoculum buildup in the soil.

## **29. Management of plant nutrients**

The plant nutrients in general when applied in excess may increase or reduce the resistance in plants to diseases. Increased application of nitrogenous fertilizers increases the incidence of many diseases. Crops fed with heavy doses of nitrogenous fertilizers grow robust with foliage and succulent tissue but become highly susceptible to the attack of diseases like rust, powdery mildew, blast, tobacco mosaic and some bacterial diseases. In the case of blast

of rice optimum dose of nitrogenous fertilizers are recommended and it is applied in 3 split doses viz. 50% as based at transplanting, 25% at tillering and 25% at panicle initiation stage. Late application of nitrogenous fertilizers increases wheat leaf blotch (*Septoria nodorum*) and powdery mildew (*Erysiphe graminis tritici*).

Some diseases are favoured by ammoniacal form of nitrogen while others are favoured by nitrate form of nitrogen. In general wilts (*Fusarium* sp.) and root rots (*Rhizoctonia* spp.) are favoured by ammoniacal nitrogen while *Verticillium* wilts and root rots due to *Pythium* spp. are favoured by nitrate nitrogen. In rice, blast disease is favoured by ammoniacal nitrogen while brown spot (*Helminthosporium oryzae*) is favoured by nitrate nitrogen. In maize Northern corn leaf blight caused by *H. turcicum* is favoured by ammoniacal nitrogen while stalk rot (*Diplodia maydis*) is favoured by nitrate nitrogen.

In wheat, sharp eye spot (*Rhizoctonia solani*) is favoured by ammoniacal nitrogen while stem rust (*Puccinia graminis tritici*) is favoured by nitrate nitrogen. In potato, wilt (*Verticillium albo-atrum*) and scab (*Streptomyces scabies*) are favoured by nitrate nitrogen while ammoniacal nitrogen increases black scurf (*R. olani*).

Effects of nitrogenous fertilizers on major soil borne diseases have been studied. Their effect on the disease i.e., whether increased or decreased incidence by nitrogen in different forms are given in the following table.

**Table. Effects of different forms of nitrogen on soil-borne diseases**

Pathogen	Host	Amendment
<b>Diseases increased</b>		
<i>Fusarium oxysporum</i> f.sp.	Tomato	NO <sub>3</sub>
<i>lycopersici</i>	Sorghum	NaNO <sub>3</sub> -
<i>F.moniliforme</i>	Carnation	NH <sub>4</sub> NO <sub>3</sub> NO <sub>3</sub>
<i>F.roseum</i>	Bean	NH <sub>4</sub> (NH <sub>4</sub> )
<i>F.solanif.sp.phaseoli</i>	Wheat	<sub>2</sub> SO <sub>4</sub> NO <sub>3</sub>
<i>Gaeumannomyces graminis</i>	Tabacco	(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> .Ca(NO <sub>3</sub> ) <sub>2</sub> .KNO <sub>3</sub>
<i>Phytophthora nicotianae</i>	Cotton	NH <sub>4</sub> NO <sub>3</sub> +CaCO <sub>3</sub>
var. <i>nicotianae</i>	Potato	
<i>Verticillium albo-atrum</i>		
<i>Streptomyces scabies</i>		



<b>Disease decreased</b>		
<i>F. oxysporum</i> f. sp. <i>cubense</i>	Banana	Urea (nitrite)
<i>F. solani</i> f. sp. <i>phaseoli</i>	Bean	KNO <sub>3</sub> (NH <sub>4</sub> )
<i>Gaeumannomyces graminis</i>	Wheat	<sub>2</sub> SO <sub>4</sub> KNO <sub>3</sub>
<i>Phytophthora cinnamomi</i>	Avocado	Ca(NO <sub>3</sub> ) <sub>2</sub>
<i>Sclerotium rolfsii</i>	Tomato	NH <sub>3</sub> ·(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> ·Ca(NO <sub>3</sub> ) <sub>2</sub>
<i>S. rolfsii</i>	Sugarbeet	

Repeated application of phosphatic fertilizers delays the onset and lessens the severity of take-all disease of barley (*Gaeumannomyces graminis*). Potassium application reduces the disease incidence in many crop diseases probably by increasing phenolics synthesis in plants. Application of potash induces resistance in groundnut against root rot caused by *Macrophomina phaseolina*. Calcium application suppresses the lesions due to the *R. solani* on bean roots. It is due to formation of calcium pectate, which is less available to action by polygalacturanase (PG) enzyme than is pectic acid.

Calcium has also been shown to affect *Sclerotium rolfsii* by neutralizing the oxalic acid produced by the fungus. Application of molybdenum reduces infection of potato tubers by *Phytophthora infestans* and also diminishes incidence of *Ascochyta* blight on beans and peas. Manganese reduces late blight of potato, ferric chloride controls rice brown spot and silicon application reduces rice blast.

### **30. Time of harvesting**

Time of harvesting affects the cleanliness of the seeds. Delayed harvesting of grain crops in temperate climatic conditions enables the pathogen more time to contaminate the seeds. The best example is grain mould of sorghum where contamination by species of *Fusarium*, *Curvularia*, *Alternaria*, *Aspergillus*, *Phoma* is seen. Potato tubers harvested when the tops are green get easily contaminated by the late blight pathogen present on the leaves. Removal of tops and making them dry before digging the tubers kills the sporangia and avoids contamination of tubers harvested later.

### **31. Avoiding ratoons**

Ratooning is a general practice in sugarcane when the incidence of grassy shoot disease and red rot are very high. Hence ratooning should be avoided.

### **32. Solarheating**

When the soil is covered with white polythene sheets during hot seasons, soil temperature increases. Increased soil temperature eliminates wilt pathogens like *Fusarium oxysporum*

f.sp. *lycopersici* and *Verticillium dahliae* from tomato field. High soil temperature also favours antagonistic fungi.

**Biological control and PGPR – Scope and importance – Role and mechanisms of biological control and PGPR with examples. Plant growth promoting rhizobacteria**

Biological control is defined as the reduction of inoculum density or disease producing activities of a pathogen or parasite in its active or dormant stage by one or more organisms accomplished naturally or through manipulation of the environment, host or by introduction of one or more antagonists or by mass introduction of one or more antagonists.

Biological control is but control of plant diseases using living microorganisms. Root rot disease (*Macrophomina phaseolina*) is a major disease in pulses, oilseeds, cotton, etc., and the most common method of control is using fungicides. But the chemical methods are uneconomical and less effective, as seed treatment with chemical may give protection only in the early stages of crop growth 2 weeks.

In addition, it is harmful to the beneficial microorganisms in soil and creates residual problems. So, the biological control can be very effectively used for the root rot disease management as the biological agent multiplies in soil and offers protection throughout the crop growth. The four main mechanisms involved in the biocontrol are (i) the biological agent (antagonist) may parasitize the other organism, (ii) antagonist may secrete metabolites (antibiotics) harmful to the pathogens (Antibiosis) (iii) antagonist may compete with the pathogens for nutrients or space (Competition) and (iv) may cause death of the parasite by producing enzymes (Lysis).

**Parasitism and Lysis**

The biocontrol agent parasitizes the pathogen by coiling around the hyphae, e.g., *Trichoderma viride*; various bacteria and fungi secrete hydrolytic enzymes about the degradation of cell walls of pathogens.

e.g. (i) *Bacillus* sp. causes hyphal lysis of *Gaeumannomyces graminis*

(ii) The chitinolytic enzymes of *Serratia marcescens* caused cell wall lysis of *Scierotium rolfsii*. (iii) *Trichoderma* sp. produces chitinases and  $\beta$ -1,3-glucanases which lyse the cell wall of *Rhizoctonia solani*.

### **Antibiosis**

The antibiotic compounds secreted by the biocontrol agent suppress the growth of the pathogen. e.g. Phenazine-1-carboxylic acid produced by *P fluorescens* plays an important role in suppressing the take-all disease of wheat.

### **Competition**

The biocontrol bacteria and fungus compete for food and essential elements with the pathogen there by displacing and suppressing the growth of pathogen.

e.g.

(i) the competition for nutrients between *Pythium aphanidermatum*, *Pultimum* and bacteria suppress the damping off disease in cucumbers.

(ii) Fluorescent siderophores (iron chelators) such as pseudobactin and pyoverdins produced by *P fluorescens* chelates iron available in the soil, thereby depriving the pathogen of its requirements.

### **A. TRICHODERMA VIRIDE**

The fungus, *Trichoderma viride* is one such biocontrol agent, mainly used for the control of root rot diseases of pulses and oil seeds in Tamil Nadu. A mass production technology for *T. viride* has been developed by Tamil Nadu Agricultural University, Coimbatore.

#### **Systematic Position**

Asexual (conidial) Sexual (ascospore)

Subdivision: Deuteromycotina Ascomycotina Cl

Class: Hypomycetes Pyrenomycetes

Order : Moniliales

Sphaeriales Family: Moniliaceae Hyp

ocreaceae Genus : Trichoderma

Hypocrea **Isolation of Trichoderma from soil**

#### **from soil**

*Trichoderma* is isolated from the soil by using *Trichoderma* selective medium developed by Elad and Chet (1983). Collect soil samples from the field, mix well and make it into fine particles. Soil samples should be collected in root zone at 5-15 cm depth and from rhizosphere wherever possible. Ten gram of soil sample is taken, and suspended in 100 ml of sterile distilled water and stirred well to get 1:10 dilution. Transfer one ml from this to 9 ml of sterile water in a test tube to get 1:100 dilution. Make serial dilutions by transferring one ml of

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suspension

to subsequent tube to get dilution of 1:10,000. Transfer one ml of the desired soil suspension to

sterile petriplates. Pour 15 ml of melted and cooled *Trichoderma* selective medium in the same petriplates. Rotate the plate gently and allow to solidify, incubate at room temperature for 5-7 days and observe for the development of fungal colonies. *Trichoderma* colonies will be white initially and turn to green. Count the number of colonies developing in individual plates. Transfer the individual colonies to potato dextrose agar slants.

### **Testing Method**

#### **Dual Culture Technique**

It consists of growing the test organism and the pathogenic organism on the same plate. This can be done by the following procedure. Transfer 15-20 ml of melted and cooled PDA to sterilised petri dishes. Allow it to solidify. Transfer 8 mm disc of test organism to one end of the petriplate. In the opposite end, 8 mm disc of the pathogenic culture is transferred in the same petriplate (if the antagonistic micro-organism is slow growing it should be plated in the previous day itself). Incubate the plate at room temperature. Observe the development of inhibition zone. Observe under microscope where both the test organism and the pathogen come in contact.

#### **Mass Production**

Molasses yeast medium (Molasses 30g + yeast 5g + water 1000ml) is prepared in conical flasks and sterilized at 1.1 kg/cm<sup>2</sup> for 20 minutes. *T. viride* culture is inoculated by taking a fungal disc from 10 day old culture and incubated for 10 days. This serves as mother culture. Molasses yeast medium is prepared in a fermenter and sterilized. Then, the mother culture is added to the fermenter @ 1.5 litre/50 litres of medium and incubated at room temperature for 10 days. The fungal biomass and broth are mixed with talc powder at 1:2 ratio. The mixture is air dried and mixed with carboxy methyl cellulose (CMC) @ 5g / kg of the product. It is packed in Polythene covers and used within 4 months.

#### **Quality Control Specifications**

1. Fresh product should contain not less than  $28 \times 10^6$  CFU/g
2. After 120 days of storage at room temperature, the population should be  $10 \times 10^6$  cfu/g.
3. Maximum storage period using talc carrier is 120 days.
4. Size of the carrier (talc) should be 500 microns.
5. Product should be packed in white Polythene bags.
6. Moisture content of the final product should not be more than 20%.

### ***B. Bacillus subtilis***

This bacterium is widely used for the control of soil-borne plant pathogens like *Macrophomina phaseolina*, *Rhizoctonia solani*, *Fusarium* spp. etc. This treatment also considerably improves the plant growth and yield. *Bacillus subtilis* is a rod shaped, thermophilic gram positive, aerobic bacterium. Spores may be formed in chains. It is 5-6  $\mu$ m in length and 2-3  $\mu$ m in width. It forms endospores during adverse conditions.

#### **Isolation**

One gram of soil sample is mixed with 9 ml sterilized nutrient broth in a test tube. This has to be kept on a boiling water bath at 80°C for 10 minutes. Then it is kept for incubation at room temperature for 24-48 hrs. From this serial dilution is prepared upto  $10^{-6}$  dilution. Dilutions  $10^{-5}$  and  $10^{-6}$  are plated in Nutrient Agar and incubated for 24-48 hrs. *B. subtilis* colonies will be rough, opaque with irregular margins.

#### **Staining for Identification**

Bacterial smear is prepared with 24 hours old culture, air dried and heat fixed. The slide is flooded with crystal violet for 60 seconds and then washed with tap water. Then, the slide is flooded with Gram's iodine mordant for 60 seconds and washed with tap water. It is then the smear is counterstained with safranin for 30 seconds, washed with tap water, blotted and observed under microscope. *Bacillus subtilis* appeared violet since it is gram positive.

#### **Biochemical tests for Identification**

The following biochemical tests are carried out for identification.

1. Starch hydrolysis
2. Catalase test
3. Nitrate reduction test
4. Acid and gas production test

*Bacillus subtilis* is amylase positive, catalase positive, nitrate positive, acid positive and gas negative.

#### **Mass multiplication**

Nutrient broth (Peptone 5g, beef extract 3g, sodium chloride 3g in 1 litre of distilled water, pH 7) is prepared and sterilized at 1.1 kg/cm<sup>2</sup> pressure for 20 minutes. One loopful of *B. subtilis* is inoculated and incubated for 24 hours. This serves as mother culture. One litre of mother culture is transferred to 100 litres of sterilized nutrient broth in a fermenter and the

bacterial growth is harvested after 72 hrs. Then it is mixed with 250 kg of sterilized peat soil amended with 37 kg Calcium carbonate, dried in shade and packed in Polythene bags. This product can be stored up to 6 months.

### **C. *Pseudomonas fluorescens***

This is another bacterium effectively used in controlling sheath blight and blast of paddy, wilt diseases of redgram, and banana. *Pseudomonas fluorescens* is a gram negative, rod shaped non-spore forming bacterium which may be mono or loptrichous or non-motile. It produces greenish, fluorescent and water soluble pigment, pyoverdinin. The direct influence of *Pseudomonas* on plant growth is mediated either by release of auxin-like substances or through improved uptake of nutrients in the environment. The indirect promotion of plant growth is achieved when fluorescent *Pseudomonas* decreases or prevents the deleterious influence of phytopathogens.

### **Isolation**

One gram of rhizosphere soil sample is mixed in 100 ml of sterile water to give 1:100 dilution. From this serial dilutions up to  $10^{-7}$  level are made by repeatedly transferring 1 ml of 1:100 dilution to 9 ml sterile water. Stants  $10^{-5}$ ,  $10^{-6}$  and  $10^{-7}$  dilutions are plated in Kings B Agar medium and incubated for 24-48 hours. *P. fluorescens* appears as smooth, slimy, circular translucent colonies.

### **Mass production**

*P. fluorescens* is multiplied in sterilized Kings 'B' broth for 48 hours. The pH of the substrate (Peat soil or talc powder) is adjusted to 7 by adding calcium carbonate @ 150g / kg. The substrate is then sterilized at 1.1 kg/cm<sup>2</sup> pressure for 30 minutes for two successive days. Four hundred ml of *P. fluorescens* suspension is added to 1 kg of substrate containing 5 g of carboxy methyl cellulose and mixed well. The formulation is packed in Polythene covers and can be stored for one month.

### **Quality Control**

1. Fresh product should contain  $2.5 \times 10^8$  cfu / g
2. After 3 months of storage at room temperature, the population should be  $8-9 \times 10^7$  CfU / g.
3. Storage period is 3-4 months
4. Minimum population load for use is  $1.0 \times 10^8$  cfu / g.
5. Product should be packed in white Polythene bags.



6. Moisture content of the product should not be more than 20% in the final product.

7. Population per ml of the broth is  $9 \pm 2 \times 10^8$  cfu / g.

### **Methods of Application**

Crop: Paddy-blast, sheath blight

#### **1. Seed Treatment**

Mix paddy seeds with the formulation at the rate of 10 g per kg of seeds and soak these seeds in water for overnight. Decant the excess water and allow to sprout the seeds for 24 hrs and then sow.

#### **2. Seedling root dipping**

Apply 2.5 kg of the formulation to the water stagnated in an area of 25 sq.m. These seedlings, after pulling out from the nursery can be left in the stagnating water containing the bacteria. A minimum period of 30 minutes is necessary for soaking the roots and prolonged soaking will enhance the efficacy.

#### **3. Soil application**

Apply the product @ 2.5 kg / ha after 30 days of transplanting (This product should be mixed with 50 kg of well decomposed FYM / sand and then applied).

#### **4. Foliar application**

Spray the product at 0.2% concentration (1 kg/ha) commencing from 45 days after transplanting at 10 days interval for 3 times depending on disease intensity. If there is no disease incidence, a single spray is sufficient. Crop: Groundnut, Gingelly, Sunflower, Red gram, Greengram, Black gram-root rot and wilt

Seed treatment: 10g/kg of seeds

Soil application: Apply 2.5 kg/ha. mixed with 50 kg of well decomposed FYM/sand at 30 days after sowing

.

### **Crop: Banana -Fusarium wilt**

Sucker treatment: 10g/sucker

Capsule application: 50mg/capsule/sucker.

Apply once in 3 months from 3 months after

planting Soil application: 2.5 kg /ha+50 kg FYM /

sand

Apply once at the time of planting and repeat it once in 3 months.

### **Plant Products and Antiviral principles in plant disease management**

Plant products play an important role in evolving an ecologically sound and environmentally acceptable disease management system. Plant products have been found to have fungicidal, bactericidal and antiviral properties. It is well established that about 346 plant products have fungicidal properties, 92 have bactericidal and 90 have antiviral properties. This clearly indicates that the plant kingdom is a vast storehouse of chemicals that can check several plant pathogens. As many of them have more than one type of activity there is a less chance for development of resistance and moreover, the plant products are safe to non-target organisms.

### **Neem Products**

Among the plant products, the neem derivatives are reported to be effective in controlling several diseases. The neem tree (*Azadirachta indica*), popularly called as china berry, crackjack, Nim, Indian lilac, margosa and paradise tree, contains several active principles in various parts. The important active principles are Azadirachtin, Nimbin, Nimbidin, Nimbinene, Nimbridic acid and Azadirone which have antifungal and insecticidal properties.

#### **(i) Neem Seed Kernel Extract (NSKE)**

It is prepared by soaking 5 kg of powdered neem seed kernel (in a gunny bag) in 100 litres of water for 8 hours. The gunny bag is then removed after thorough shaking. Then, 100 ml of teepol is mixed thoroughly, before spraying. The quantity of extract required for a hectare is 500 litres,

#### **(ii) Neem oil solution**

One hundred ml of teepol is mixed first with 100 litres of water. Then, 3 litres of neem oil is slowly added to this solution with constant shaking. The milky solution formed is ready for spray. The spray volume is 500 litres/ha.

#### **(iii) Neem cake extract**

Ten kg of powdered neem cake in a gunny bag is soaked in 100 litres of water for 8 hours. The gunny bag is removed after thorough shaking. Then, 100 ml of sticker is added and mixed well. The quantity of spray fluid required is 500 litres/ha.

#### **(iv) Neem cake**

Powdered neem cake is directly applied to the field at the time of last ploughing. The quantity applied is 150 kg/ha.

### **Diseases controlled by neem products**

**(a) Paddy: Tungro (virus)** (Vector: *Nephotettix virescens*)

Neem cake is applied at 150 kg/ha as basal dose. In addition, 3% neem oil or 5% NSKE @ 500 l/ha can be sprayed. If one jassid is noticed in a plant. Three sprays have to be given at 15 days interval.

**(b) Paddy: Sheath rot (*Acrocyfndrium oryzae*)**

Five per cent NSKE or 3% neem oil can be sprayed @ 500 lit/ha at the time of grain emergence.

**(c) Paddy: Blast (*Pyricularia oryzae*)** Spraying 5% neem oil is effective

**(d) Paddy: Sheath blight (*Rhizoctonia solani*)**

Application of 150 Kg of neem cake/ha

**(e) Groundnut: Rust (*Puccinia arachidis*)**

Application of 3% neem oil @ 500 lit/ha. The first sprays should be given immediately on noticing the symptom and second 15 days later.

**(f) Groundnut: Foot rot (*Sclerotium rolfsii*)** Application of 1% neem oil is effective.

**(g) Coconut: Wilt (*Ganoderma lucidum*)**

Application of 5 kg of neem cake/tree/year during the rainy season.

**(h) Blackgram: Powdery mildew (*Erysiphe polygoni*)**

Two sprays with 3% neem oil or 5% NSKE, starting first spray at the initiation of the disease and second 15 days later are effective.

**(i) Blackgram: Root rot (*Macrophomina phaseolina*)** Application of neem cake @ 150 kg/ha

**(j) Blackgram: Yellow mosaic (Virus)** Application of 3% neem oil is effective.

**(k) Soybean: Root rot (*M. phaseolina*)** Application of neem cake @ 150 kg/ha.

### **Other Plant Products**

In addition to the neem products, products from several other plant species are also found to be effective in disease management. The leaf extract of tuisi (*Ocimum sanctum*) is found effective against *Helminthosporium oryzae* (paddy brown spot). The leaf and pollen extracts of vilvam (*Aegle marmelos*) effectively reduced early blight of tomato (*Alternaria solani*) and blight of onion (*A. porri*). *A. solani* is also effectively checked by flower extract of periwinkle (*Catharanthus roseus*) and bulb extract of garlic (*Allium sativum*).

Rice discolouration caused by *Drechslera oryzae* is effectively reduced by leaf extract of mint (*Mentha piperita*). The bulb extract of garlic is also effective in reducing leaf blight of finger millet (*H. nodulosum*) and blast of paddy (*Pyricularia oryzae*). The root exudates of kolinji and rhizome extract of banana are effectively used against *Ganoderma lucidum*, the pathogen of Thanjavur wilt of coconut. The seed oil of pinna (*Calophyllum inophyllum*) is effective against *Puccinia arachidis* causing groundnut rust. Leaf extract of nochi (*Vitex negundo*) effectively reduced Rice Tungro viruses by checking the vector, *Nephotettix virescens*.

### **Anti Viral Principle (AVP)**

Plants are also known to contain some compounds which are inhibitory to virus. They are called Anti-Viral Principles (AVP) or Anti Viral Factors (AVF). The leaf extracts of sorghum, coconut, bougainvillea, *Prosopis juliflora* and *Cyanodon dactylon* are known to contain virus inhibiting principles.

### **Preparation of AVP extract**

Dried coconut or sorghum leaves are cut and powdered. Twenty kg of leaf powder is mixed with 50 litres of water and heated at 60 °C for one hour. It is filtered and volume is made up to 200 litres. This gives 10 per cent extract. Five hundred litres of extract is required to cover one hectare. The 10 per cent AVP extract is very effective in controlling groundnut ring mosaic virus (bud necrosis).

Two sprays are to be given at ten and twenty days after sowing. Similarly 10 per cent leaf extracts of *P. juliflora* and *C. dactylon* effectively reduced the tomato spotted wilt virus in tomato. The leaf extracts are known to contain some proteinaceous substances which induce virus inhibition in the plants.

### **PGPR**

Plant growth promoting rhizobacteria are bacteria that colonize plant roots, and in doing so, they promote plant growth and/or reduce disease or insect damage. There has been much research interest in PGPR and there is now an increasing number of PGPR being commercialized for crops. Organic growers may have been promoting these bacteria without knowing it. The addition of compost and compost teas promote existing PGPR and may introduce additional helpful bacteria to the field. The absence of pesticides and the more complex organic rotations likely promote existing populations of these beneficial bacteria. However, it is also possible to

inoculate seeds with bacteria that increase the availability of nutrients, including solubilizing phosphate, potassium, oxidizing sulphur, fixing nitrogen, chelating iron and copper. Phosphorus

(P) frequently limits crop growth in organic production. Nitrogen fixing bacteria are miniature of urea factories, turning N<sub>2</sub> gas from the atmosphere into plant available amines and ammonium via a specific and unique enzyme they possess called nitrogenase. Although there are many bacteria in the soil that 'cycle' nitrogen from organic material, it is only this small group of specialized nitrogen fixing bacteria that can 'fix' atmospheric nitrogen in the soil. Arbuscular mycorrhizal fungi (AMF) are root symbiotic fungi improving plant stress resistance to abiotic factors such as phosphorus deficiency or desiccation.

The fourth major plant nutrient after N, P and K is sulphur (S). Although elemental sulphur, gypsum and other sulphur bearing mined minerals are approved for organic production, the sulphur must be transformed (or oxidized) by bacteria into sulphate before it is available

for plants. Special groups of microorganisms can make sulphur more available, and do occur naturally in most soils.

One of the most common ways that PGPR improve nutrient uptake for plants is by altering plant hormone levels. This changes root growth and shape by increasing root branching, root mass, root length, and/or the amount of root hairs. This leads to greater root surface area, which in turn, helps it to absorb more nutrients.

### **Disease control**

PGPR have attracted much attention in their role in reducing plant diseases. Although the full potential has not been reached yet, the work to date is very promising and may offer organic growers some of their first effective control of serious plant diseases. Some PGPR, especially if they are inoculated on the seed before planting, are able to establish themselves on the

crop roots. They use scarce resources, and thereby prevent or limit the growth of pathogenic microorganisms. Even if nutrients are not limiting, the establishment of benign or beneficial organisms on the roots limits the chance that a pathogenic organism that arrives later will find space to become established. Numerous rhizosphere organisms are capable of producing compounds that are toxic to pathogens like HCN

### **Challenges with PGPR**

***Epidemiology & Principles of DM***

One of the challenges of using PGPR is natural variation. It is difficult to predict how an organism may respond when placed in the field (compared to the controlled environment of a

laboratory. Another challenge is that PGPR are living organisms. They must be able to be propagated artificially and produced in a manner to optimize their viability and biological activity until field application. Like Rhizobia, PGPR bacteria will not live forever in a soil, and over time growers will need to re-inoculate seeds to bring back populations.

### **PGPR in Research**

Over the years the PGPR (plant growth promoting rhizobacteria) have gained worldwide importance and acceptance for agricultural benefits. These microorganisms are the potential tools for sustainable agriculture and the trend for the future. Scientific researchers involve multidisciplinary approaches to understand adaptation of PGPR to the rhizosphere, mechanisms of root colonization, effects of plant physiology and growth, biofertilization, induced systemic resistance, biocontrol of plant pathogens, production of determinants etc. Biodiversity of PGPR and mechanisms of action for the different groups: diazotrophs, bacilli, pseudomonads, Trichoderma, AMF, rhizobia, Phosphate solubilising bacteria and fungi, Lignin degrading, chitin degrading, cellulose degrading bacteria and fungi are shown. Effects of physical, chemical and biological factors on root colonization and the proteomics perspective on biocontrol and plant defense have also shown positive results. Visualization of interactions of pathogens and biocontrol agents on plant roots using autofluorescent protein makers has provided more understanding of biocontrol processes with overall positive consequences.

### **Ways that PGPR promote plant growth**

- Increasing nitrogen fixation in legumes
- Promoting free-living nitrogen-fixing bacteria
- Increasing supply of other nutrients, such as phosphorus, sulphur, iron and copper
- Producing plant hormones
- Enhancing other beneficial bacteria or fungi
- Controlling fungal diseases
- Controlling bacterial diseases
- Controlling insect pests

## **Lecture 08**

### **Physical Methods–**

### **Heat treatments, soil solarization, hot water treatment, hot air treatment, control by refrigeration and radiation**

As early as 1832, Sinclair suggested that hot air treatment in an oven might control smut of oats and barley. Gardeners in Scotland while treating the bulbs of different ornamental plants first employed hot water therapy.

The scientific principle involved in heat therapy is that the pathogen present in seed material is selectively inactivated or eliminated at temperatures that are non-lethal to the host tissues.

Following physical methods are employed for reduction or elimination of primary inoculum that may be present in seed, soil or planting material.

#### **i. Hot water treatment (HWT)**

The seeds are soaked in cold water at 20-30°C for 5 hrs to induce the dormant mycelium to grow. Then the seeds are immersed in hot water at 50-54°C for 10 minutes to kill the mycelium. It is very effectively used to eliminate loose smut of wheat. The sets of sugarcane can be treated at 50°C for 2 hrs to eliminate grassy shoot pathogen. The main drawback in the hot water treatment is that the seeds may be killed or lose their germinability, if the period of treatment exceeds the specified time. So this method is replaced by other physical methods like hot air and aerated steam treatment wherein the seeds are exposed only to hot air/aerated steam.

#### **ii. Hot air treatment (HAT)**

Sugarcane sets are treated with hot air at 50°C for 2 hrs to eliminate mosaic virus.

#### **iii. Aerated steam therapy (AST)**

Sugarcane sets are also exposed to aerated steam at 50°C for 3 hrs to eliminate mosaic virus.

#### **iv. Moist hot air treatment (MHAT)**

This method is effectively used in sugarcane to eliminate grassy shoot disease. Initially the sets are exposed to hot air at 54°C for 8 hrs, then exposed to aerated steam at 50°C for 1 hr and finally to moist hot air at 54°C for 2 hours.

#### **v. Solar heat treatment (SHT)**

A simplest treatment has been devised in India to eliminate the pathogen of loose smut of wheat. Previously the hot water treatment was followed to eliminate loose smut. As the thermal



death point of the fungus and the embryo are very close. The extensive care should be taken to avoid killing of the embryo. Luthra in 1953 devised a method to eliminate the deep seated infection of *Fusarium*. The method is popularly known as solar heaters or solar energy treatment.

Luthra's solar energy treatment: The seeds are soaked in cold water for 4 hours in the forenoon on a bright summer day followed by spreading and drying the seeds in hot sun for four hours in the afternoon. Then, the seeds are again treated with carboxin or carbendazim at 2g/kg and stored. This method is highly useful for treating large quantities of these seed lots.

#### **vi. Soil Solarization**

Soil solarization is generally used for controlling soil-borne pathogens like *Pythium*, *Verticillium*, *Rhizoctonia*, *Fusarium* etc. and nematodes in small areas like nurseries. Irrigate the nursery bed to moisten the soil to a depth of 10cm. Cover the bed after 2 days with thin transparent polyethylene sheets for 4-6 weeks and then irrigate the beds once in a week. The purpose of irrigation is to increase the thermal sensitivity of resting structures of fungi and to improve heat conduction.

#### **vii. Steam Sterilization**

Steam is passed through perforated pipes at a depth of 15 cm to sterilize the upper layers of soil. It is mostly practiced under glass house and green house conditions.

#### **viii. Hot Air Sterilization**

Hot air is also passed through pipelines to sterilize the soils in the nursery areas.

#### **ix. Hot Water Treatment**

It is mainly done in pot culture studies to kill the fungi and nematodes. The pots containing soil are immersed in boiling water at 98°C for 5 minutes or drenching boiling water @ 20 litres/ Sq.m.

#### **Refrigeration**

It is an accepted fact that the low temperature at or slightly above the freezing point checks the growth and activities of all such pathogens that cause a variety of post harvest diseases of vegetables and fruits. Therefore most perishable fruits and vegetables should be transported and stored in refrigerated vehicles and stores. Cool chains refrigerated space from field to consumer table is becoming very popular. Regular refrigeration is sometimes preceded

by a quick hydrocooling or air cooling to remove the excess heat carried in them from the field to prevent development of new or latent infections.

### **Radiation**

Electromagnetic radiations such as ultraviolet (UV) light, x rays and y rays as well as particulate radiations have been studied in relation to management of post harvest diseases of horticultural crops. Y rays controlled post harvest fungal infections in peaches, straw berries and tomatoes but doses of radiation required to kill pathogens, were found injurious to host tissues. Some plant pathogenic fungi sporulate only when they receive light in the ultraviolet range. It has been possible to control diseases on green house vegetables caused by species of these fungi by covering or constructing the green house with a special UV absorbing vinyl film that blocks transmission of light wavelengths below 390 nm.

**Chemical methods—study of different groups of fungicides.**

**Methods of application of fungicides**

**Fungicides—definition**

The word „fungicide“ originated from two Latin words, viz., „fungus“ and „caedo“. The word „caedo“ means „to kill.“ Thus the fungicide is any agency/chemical which has the ability to kill the fungus. According to this meaning, physical agents like ultra violet light and heat should also be considered as fungicides. However, in common usage, the meaning is restricted to chemical only. Hence, fungicide is a chemical which is capable of killing fungi.

**Fungistat**

Some chemicals do not kill the fungal pathogens. But they simply arrest the growth of the fungus temporarily. These chemicals are called fungistat and the phenomenon of temporarily inhibiting the fungal growth is termed as fungistat.

**Antisporulant**

Some other chemicals may inhibit the spore production without affecting the growth of vegetative hyphae and are called as „Antisporulant“. Even though, the antisporulant and fungistat compounds do not kill the fungi, they are included under the broad term fungicide because by common usage, the fungicide has been defined as a chemical agent which has the ability to reduce or prevent the damage caused to plants and their products. So, some of the plant pathologists prefer the term „Fungitoxicant“ instead of fungicide.

**Characters of an ideal fungicide**

1. It should have low phytotoxicity
2. It should have long shelf life
3. Stability during dilution
4. It should be less toxic to human being, cattle, earthworms, microorganisms etc.
5. It should be a broad spectrum insecticide
6. Fungicide preparation should be ready for use
7. It should have compatibility with other agrochemicals
8. It must be cheaper
9. It should be available in different formulations
10. It should be easily transportable

## **Classification of Fungicides**

Fungicides can be broadly grouped based on their (i) mode of action (ii) general use and (iii) chemical composition.

### **I. Based on mode of action**

**Protectant**  
As the name suggests, protectant fungicides are prophylactic in their behaviour. Fungicide which is effective only if applied prior to fungal infection is called a protectant, eg., Zineb, Sulphur.

### **Therapeutant**

Fungicide which is capable of eradicating a fungus after it has caused infection and thereby curing the plant is called chemotherapeutant. eg. Carboxin, Oxycarboxin antibiotics like Aureofungin. Usually chemo therapeutant are systemic in their action and affect the deep-seated infection.

### **Eradicant**

Eradicant are those which remove pathogenic fungi from an infection court (area of the host around a propagating unit of a fungus in which infection could possibly occur). eg. Organic mercurials, lime sulphur, dodine etc. These chemicals eradicate the dormant or active pathogen from the host. They can remain effective on or in the host for some time.

### **II. Based on general uses**

The fungicides can also be classified based on the nature of their use in managing the diseases.

1. Seed protectants: Eg. Captan, thiram, organomercurials, carbendazim, carboxin etc.
2. Soil fungicides (preplant): Eg. Bordeaux mixture, copper oxychloride, Chloropicrin, Formaldehyde Vapam, etc.,
3. Soil fungicides: Eg. Bordeaux mixture, copper oxy (for growing plants) chloride, Captan, PCNB, thiram etc.
4. Foliage and blossom: Eg. Captan, ferbam, zineb, protectants mancozeb, chlorothalonil etc.
5. Fruit protectants: Eg. Captan, maneb, carbendazim, mancozeb etc.
6. Eradicants: Eg. Organomercurials, lime sulphur, etc.
7. Tree wound dressers: Eg. Bordeaux paste, chaubatti paste, etc.

8. Antibiotics: Eg. Actidione, Griseofulvin, Streptomycin, Streptocycline, etc.,
9. General purposes spray and dust formulations.

### **III. Based on Chemical Composition**

The chemical available for plant disease control runs into hundreds, however, all are not equally safe, effective and popular. Major group of fungicides used include salts of toxic metals and organic acids, organic compounds of sulphur and mercury, quinines and heterocyclic nitrogenous compounds. Copper, mercury, zinc, tin and nickel are some of the metals used as base for inorganic and organic fungicides. The non metal substances include, sulphur, chlorine, phosphorus etc. The fungicides can be broadly grouped as follows and discussed in detail.

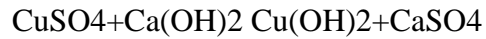
#### **Group of Fungicides – Copper Fungicides, Sulphur Fungicides and Mercury Fungicides**

The fungicidal action of copper was mentioned as early as 1807 by Prevost against wheat bunt disease (*Tilletia caries*), but its large scale use as a fungicide started in 1885 after the discovery of Bordeaux mixture by Millardet in France. The mixture of copper sulphate and lime was effective in controlling downy mildew of grapevine caused by *Plasmopara viticola* and later, late blight of potato (*Phytophthora infestans*).

Some other copper sulphate preparations later developed were Bordeaux paste, Burgandy mixture and Cheshnut compound which are all very effectively used in the control of several plant diseases. In addition some preparations of copper oxy chloride preparations are also used. These are all insoluble copper compounds very successfully used in managing several leaf diseases and seedling diseases in nursery. Some of the important diseases controlled by copper fungicides are listed below.

#### **I. Copper sulphate preparations Bordeaux Mixture**

In 1882, Millardet in France (Bordeaux University) accidentally observed the efficacy of the copper sulphate against the downy mildew of grapes caused by *Plasmopara viticola*. When copper sulphate was mixed with lime suspension, it effectively checked the disease incidence. The mixture of copper sulphate and lime was named as “Bouillie Bordelaise” (Bordeaux Mixture). The original formula developed by Millardet contains 5 lbs of  $\text{CuSO}_4$  + 5 lbs of lime + 50 gallons of water. The chemistry of Bordeaux mixture is complex and the suggested reaction is:



The ultimate mixture contains a gelatinous precipitate of copper hydroxide and calcium sulphate, which is usually sky blue in colour. Cupric hydroxide is the active principle and is toxic to fungal spores. In metric system, to prepare one percent Bordeaux mixture the following procedure is adopted:

One kg of copper sulphate is powdered and dissolved in 50 litres of water. Similarly, 1 kg of lime is powdered and dissolved in another 50 litres of water. Then copper sulphate solution is slowly added to lime solution with constant stirring or alternatively, both the solutions may be prepared simultaneously to a third container and mixed well.

The ratio of copper sulphate to lime solution determines the pH of the mixture. The mixture prepared in the above said ratio gives neutral or alkaline mixture. If the quality of the used is inferior, the mixture may become acidic. If the mixture is acidic, it contains free copper which is highly phytotoxic resulting in scorching of the plants. Therefore, it is highly essential to test the presence of free copper in the mixture before applied. There are several methods to test the neutrality of the mixture, which are indicated below:

- (i) **Field Test:** Dip a well polished knife or sickle in the mixture for few minutes. If reddish deposit appears on the knife/sickle, it indicates the acidic nature of the mixture.
- (ii) **Litmus paper test:** The colour of blue litmus paper must not change when dipped in the mixture.
- (iii) **pH paper test:** If the paper is dipped in the mixture, it should show neutral pH.
- (iv) **Chemical test:** Acid a few drops of the mixture into a test tube containing 5 ml of 10% potassium ferrocyanide. If red precipitate appears, it indicates the acidic nature of the mixture.

If the prepared mixture is in the acidic range, it can be brought to neutral or near alkaline condition by adding some more lime solution into the mixture. Bordeaux mixture preparation is cumbersome and the following precautions are needed during preparation and application.

- (i) The solution should be prepared in earthen or wooden or plastic vessels. Avoid using metal containers for the preparation, as it is corrosive to metallic vessels.
- (ii) Always copper sulphate solution should be added to the lime solution, reverse the addition leads to precipitation of copper and resulting suspension is least toxic.

(iii) Bordeaux mixture should be prepared fresh every time before spraying. In case, the mixture has to be stored for a short time or a day, jaggery can be added at the rate of 100kg/100 litres of the mixture.

(iv) Bordeaux mixture is sometimes phytotoxic to apples, peaches, rice varieties like IR8 and maize varieties like Ganga Hybrid 3.

### **Bordeaux paste**

Bordeaux Paste consists of same constituents as that of Bordeaux mixture, but it is in the form of a paste as the quantity of water used is too little. It is nothing but 10 percent Bordeaux mixture and is prepared by mixing 1 kg of copper sulphate and 1 kg of lime in 10 litres of water. The method of mixing solution is similar to that of Bordeaux mixture. It is a wound dresser and used to protect the wounded portions, cut ends of trees etc., against the infection by fungal pathogens.

### **Burgundy mixture**

It is prepared in the same way as Bordeaux mixture, except the lime is substituted by sodium carbonate. So it is called as „Soda Bordeaux“. It was developed in Burgundy (France) in 1887 by Mason. The usual formula contains 1 kg of copper sulphate and 1 kg of sodium carbonate in 100 litres of water. It is a good substitute for Bordeaux mixture and used in copper-sensitive crops.

### **Cheshunt compound**

It is a compound usually prepared by mixing 2 parts of copper sulphate and 11 parts of ammonium carbonate. This formula was suggested by Bewley in the year 1921. The two salts are well powdered, mixed thoroughly and stored in an air tight container for 24 hours before being used. The ripened mixture is used by dissolving it in water at the rate of 3 g/litre. The mixture is dissolved initially in a little hot water and volume is made up with cold water and used for spraying.

## **II. Copper carbonate preparation**

### **Chaubattia Paste**

Chaubattia paste is another wound dressing fungicide developed by Singh in 1942 at Government Fruit Research Station, Chaubattia in the Almora district of Uttar Pradesh. It is usually prepared in glass containers or chinaware pot, by mixing 800g of copper carbonate and 800g of red lead in 1 litre of raw linseed oil or lanolin. This paste is usually applied to pruned parts

of apple, pear and peach to control several diseases. The paste has the added advantage that it is not easily washed away by rain water.

**III. Copper carbonate preparation**

<p>III. Cuprous oxide Preparation</p>	<p>Fungimar, Perenox, Copper Sandoz, Copper 4% dust, Percot, Cuproxid, Kirticoper.</p>	<p>Cuprous oxide is a protective fungicide, used mainly for seed treatment and for foliage application against blight, downy mildew and rusts.</p>
<p>IV. Cuperoxychloride Preparation.</p>	<p>Blitox, Cupramar 50% WP, Fytolan, Bilmix 4%, Micop D-06, Micopw-50, Bluecopper 50, Cupravit, Cobox, Cuprax, Mycop.</p>	<p>It is a protective fungicide, controls <i>Phytophthora infestans</i> on potatoes and several leaf spot and leaf blight pathogens in field.</p>

**Sulphur fungicides**

Use of sulphur in plant disease control is probably the oldest one and can be classified as inorganic sulphur and organic sulphur. Inorganic sulphur is used in the form of elemental sulphur or as lime sulphur. Elemental sulphur can be either used as dust or wettable sulphur, later being more widely used in plant disease control. Sulphur is best known for its effectiveness against powdery mildew of many plants, but also effective against certain rusts, leaf blights and fruit diseases.

Sulphur fungicides emit sufficient vapour to prevent the growth of the fungal spores at a distance from the area of deposition. This is an added advantage in sulphur fungicides as compared to other fungitoxicants.



Organic compounds of sulphur are now widely used in these days. All these compounds, called as „carbamate fungicides“, are derivatives of Dithiocarbamic acid, Dithiocarbamates are broadly grouped into two, based on the mechanism of action.

**Dithiocarbamates**

Monoalkyl Dithiocarbamates Eg. Zineb, Maneb, Eg. Thiram, Ziram, Mancozeb, Nabam, Vapam, Ferbam	Dialkyl Dithiocarbamates
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List of sulphur fungicides and the important diseases controlled by them are tabulated below:

Trade Name	Diseases Controlled	
<b>Inorganic Sulphur</b> 1. Elemental Sulphur (i) Sulphur dust	Sulphur dust Cosan, Wetsulf, Microsul	Sulphur is a contact and protective fungicide, normally applied as spray or as dust. It is generally used to control powdery mildews of fruits, vegetables, flowers and tobacco. This is also effective against apple scab ( <i>Venturia inaequalis</i> ) and rusts of field crops.
2. Lime Sulphur (Calcium polysulphide)	It can be prepared by boiling 9 Kg of rock lime and 6.75 Kg of sulphur in 225 litres of water.	Lime Sulphur is effective against powdery mildew as a protective fungicide.
<b>Organic Sulphur</b>	Hexathane 75% WP,	It is used to protect

(Dithiocarbamates) a. Monoalkyl	Dithane Z-78, Funjeb, Lonocol, Parzate C,	foliage and fruits of a wider range of crops
dithiocarbamate 1. Zineb (Zinc ethylene bisdithiocarbamate)	Du Pant Fungicide A, P olyram.	against diseases such as early and late blight of potato and tomato, downy mildews and rusts of cereals, blast of rice, fruit rot of chilli etc.
2. Maneb (Manganese ethylene bisdithiocarbamate)	Dithane M22, Manzate WP, MEB	These two are protective fungicides used to control many fungal diseases of field crops, fruits, nuts, ornamentals and vegetables, especially blights of potatoes and tomatoes, downy mildews of vines, anthracnose of vegetables and rusts of pulses.
3. Mancozeb (Maneb + Zincion)	Dithane M45, Indofil M45, Manzeb.	
4. Nabam (DSE) (DiSodium methylene bisdithiocarbamate)	Chembam, Dithane A-40, Dithane D-14, Parzate Liquid	Nabam is primarily used for foliar application against leaf spot pathogen of fruits and vegetables. Soil

		<p>applications were also reported to have a systemic action on <i>Pythium</i>, <i>Flusarium</i> and <i>Phytophthora</i>. It is also used to control algae in paddy fields.</p>
<p>5. Vapam (SMDC) (Sodium methyl dithiocarbamate)</p>	<p>Vapam, VPM, Chemvape, 4-S-Karbation, Vita Fume.</p>	<p>It is a soil fungicide and nematocide with fumigant action. It is also reported to have insecticidal and herbicidal properties. It is effective against damping off disease of papaya and vegetables and wilt of cotton. It is also effective against nematode infestation in citrus, potato and root knot nematodes in vegetables.</p>
<p><b>b. Dialkyl Dithiocarbamate</b> 1. Ziram (Zinc dimethyl dithiocarbamate)</p>	<p>Cuman L. Ziram, Ziride 80 WDP, Hexaazir 80% WP, Corozate, Fukiazsin, Karbam white, Milbam, Vancide 51 Z, Zerlate, Ziram, Ziberk, Zitox 80% WDP.</p>	<p>Ziram is a protective fungicide for use on fruit and vegetables crops against fungal pathogens including apple scab. It is non-phytotoxic except to zinc-sensitive plants. It is highly effective</p>

		against anthracnose of
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		beans, pulses, tobacco & tomato, and also against rusts of beans etc.
2. Ferbam (Ferric dimethyl dithiocarbamate)	Coromat, Febam, Ferberk, Femate, Fermate D, Fermicide, Hexaferb 75% WP, Karbam Black, Ferradow.	Ferbam is mainly used for the protection of foliage against fungal pathogens of fruits and vegetables including <i>Taphrina deformans</i> of peaches, anthracnose of citrus, downy mildew of tobacco and apple scab.
3. Thiram (Tetramethyl thiram disulphide)	Thiride 75 WDP, Thiride 750, Thiram 75% WDP, Hexathir, Normerson, Panoram 75, Thiram, TMTD, Arasan, Tersan 75, Thylate, Pomarsol, Thiuram.	It is used for seed treatment both as dry powder or as a slurry. It is a protective fungicide also suitable for application to foliage to control <i>Botrytis</i> spp. on lettuce, ornamental, soft fruits and vegetables, rust on ornamentals and <i>Venturia pirina</i> on pears. It is also effective against soilborne pathogens like <i>Pythium</i> , <i>Rhizoctonia</i> and <i>Fusarium</i> .

### **Mercury Fungicides**

Mercury fungicides can be grouped as inorganic and organic mercury compounds. Both the groups are highly fungitoxic and were extensively used as seed treatment chemicals against seed-borne diseases. Ignorance compounds show bactericidal property also. However, due to their residual toxicity in soil and plants and their extreme toxicity nature to animal and human beings, the use of mercury fungicides is being discouraged. In most of the countries, the use of mercury fungicides is banned and in countries like India, the use of mercury fungicides is restricted only in seed treatment for certain crops. The list of diseases against which mercury fungicides used are listed below

<b>Common Name</b>	<b>Trade Name</b>	<b>Diseases Controlled</b>
<b>I. Inorganic Mercury</b>		
1. Mercuric chloride	Merfusan, Mersil	It is used for treating potato tubers and propagative materials of other root crops
2. Mercurous chloride	Cyclosan, M-CTurf fungicide.	Mercurous chloride is limited to soil application in crop protection use because of its phytotoxicity.
<b>II. Organomercurials</b>	Agallol, Aretan, Emisan,	These are used mainly for treatment of seeds and planting materials. These fungicides are used for seed treatment by dry, wet or slurry method. For
Met	Ceresan wet (India)	seed treatment 1% metallic
ethoxy ethyl mercury chloride	Ceresan Dry (India), Ceresol,	mercury is applied at 0.25% concentration
Phenylmercury chloride	Leytosan.	

EthylMercuryChloride	Ceresan(USA)	
Tolylmercuryacetate	AgrosanGN.	

**Heterocyclic Nitrogen Compounds, Quinones and Miscellaneous Fungicides**

Heterocyclic nitrogen compounds are mostly used as foliage and fruits protectants. Some compounds are very effectively used as seed dressers. Some of the commonly used fungicides are listed below.

<b>CommonName</b>	<b>TradeName</b>	<b>DiseasesControlled</b>
1. Captan (Kittleson's Killer) (N-trichloromethyl thio-4-cyclohexene-1,2-dicarboximide)	Captan 50W, Captan 75W, Ezzo Fungicide 406, Orthocide 406, Vancide 89, Deltan, Merpan, Hexacap.	It is a seed dressing fungicide used to control diseases of many fruits, ornamental and vegetable crops against rots and damping off.
2. Captafol (Cis-N-1,1,2,2-tetrachlorohexane 1,2-dicarboximide)	Foltaf, Difolaton, Difosan, Captaspor, Foleid, Sanspor.	It is a protective fungicide, widely used to control foliage and fruit diseases of tomatoes, coffee and potato.
3. Glyodin	Glyoxaliadine, Glyoxide,	It has a narrow spectrum of

	Glyodin, Glyoxide Dry, Glyodex 30% liquid and 70% WP.	activity. As a spray, it controls apple scab and cherry leaf spot.
4. Folpet (Folpet) [N-(trichloromethyl-thi)] phthalimide	Phartan, Acryptan, Phaltan, Folpan, Orthophaltan.	It is also a protective fungicide used mainly for foliage application against leaf spots, downy and powdery mildew of many crops.

**Benzenecompounds**

Many aromatic compounds have important antimicrobial properties and have been developed as fungicides. Some important benzenecompounds commonly used in plant disease control are listed below.

<b>Common Name</b>	<b>Trade Name</b>	<b>Diseases Controlled</b>
1. Quintozene (PCNB)	Brassicol, Terraclor, Tritisan 10%, 20%, 40% D and 75% WP, PCNB 75% WP.	It is used for seed and soil treatment. It is effective against <i>Botrytis</i> , <i>Sclerotium</i> , <i>Rhizoctonia</i> and <i>Sclerotinia spp.</i>
2. Dichloran	Botran 50% WP and 75% WP, Allisan.	It is a protective fungicide and very effective against <i>Botrytis</i> , <i>Rhizopus</i> and <i>Sclerotinia</i> spp.



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3. Fenaminsosuph (Sodium dimethylaminobenzenediazosulphonate)	Dexon 5% G and 70% WP	It is very specific in protecting germinating seeds and growing plants from seeds as well as soil-borne infection of
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		<i>Phythium</i> , <i>Aphanomyces</i> and <i>Phytophthora</i> spp.
4. Dinocap (2,4-dinitro-6-octylphenylcrotonate)	Karathane, Arathane, DNOPC, Mildex, Crotothane, Crotothane 25% WP, Crotothane 48% Liq.	It is a non-systemic acaricide and control fungicide recommended to control powdery mildews on various fruits and ornamentals. It is also used for seed treatment.

**Quinone Fungicides**

Quinone are present naturally in plants and animals and they exhibit anti-microbial activity and some compounds are successfully developed and used in the plant disease control. Quinones are very effectively used for seed treatment and two commonly used fungicides are listed below:

<b>Common Name</b>	<b>Trade Name</b>	<b>Diseases Controlled</b>
1. Chloranil (2,3,5,6-tetrachloro-1,4-benzoquinone)	Spergon	Chloronil is mainly used as a seed protectant against smut of barley and sorghum and bunt of wheat.
2. Dichlone (2,3-dichloro-1,4-naphthoquinone)	Phygon, Phygon XL WP.	Dichlone has been used widely as a seed protectant. This is also used as a foliage fungicide, particularly against apple scab and peach leaf curl.

**Organo- Phosphorous**

**It has aspecific action**

<p><b>fungicide</b> Ediphenphos (Edifenphos)(O-ethyl-SS- diphenyldithiophosphate)</p>	<p>Hinosan 50% EC and 2% D.</p>	<p>against <i>Pyricularia oryzae</i> (Rice blast). It is also effective against <i>Corticium sesakii</i> and <i>Cochliobolus miyabeanus</i> in rice.</p>
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### Organo Tin compounds

Several other organic compounds containing tin, lead, etc. have been developed and successfully used in plant disease control. Among them, organo tin compounds are more popular and effective against many fungal diseases. These compounds also show antibactericidal properties. Some of the organo tin compounds commonly used are listed below.

Common Name	Trade Name	Diseases Controlled
<p>1. Fentin (Triphenyltin hydroxide) (Triphenyltin hydroxide)</p>	<p>Du-Ter WP 20% or 50% WP, Du-Ter Extra-WP, Farmatin 50 WP, Du-Ter forte WP, Tubotin.</p>	<p>It is a non-systemic fungicide recommended for the control of early and late blight of potato, leaf spot of sugar beet, blast of rice and tikka leaf spot of ground nut.</p>
<p>2. Fentin (Triphenyltin acetate) (Triphenyltin acetate)</p>	<p>Brestan WP 40% and 60% WP.</p>	<p>It is a non-systemic fungicide recommended to control <i>Ramularia</i> spp. on cereals and sugar beet anthracnose and downy mildew.</p> <p>It is effective against <i>Cercospora</i> leaf spot of potato.</p>

3. Fentin Chloride (TPTC-Triphenyltin chloride)	Brestanol 45% WP, Tinmate.	sugarbeet and paddy blast
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**Systemic Fungicides and Antibiotics Systemic Fungicides**

Since the late 1960s there has been substantial development in systemic fungicides. Any compound capable of being freely translocated after penetrating the plant is called systemic. A systemic fungicide is defined as fungitoxic compound that controls a fungal pathogen remote from the point of application, and that can be detected and identified. Thus, a systemic fungicide could eradicate established infection and protect the new parts of the plant.

Several systemic fungicides have been used as seed dressing to eliminate seed infection. These chemicals, however, have not been very successful in the cases of trees and shrubs.

On the basis of chemical structure, systemic fungicides can be classified as Benzimidazoles, Thiophanates, Oxathilins and related compounds, pyrimidines, morpholines, organo-phosphorus compounds and miscellaneous group.

**I. Oxathilin and related compounds**

Oxathilins were the earliest developed compounds. This group of systemic fungicide is also called as carboxamides, carboxyluc acid anillides, carboxaanillides or simply as anillides which are effective only against the fungi belong to *Basidiomycotina* and *Rhizoctonia solani*. Some of the chemicals developed are (i) Carboxin (DMOC: 5,6-dihydro-2-methyl-1,4-oxathin-3-carboxanillide) and (ii) Oxycarboxin (DCMOD-2,3-dihydro-5-carboxanillido-6-methyl-1,4-oxathilin-4,4-dioxide). The diseases controlled by these chemicals are listed below.

Common Name	Trade Name	Diseases Controlled
1. Carboxin (5,6-dihydro-2-methyl-1,4-oxanthin-3-carboxanillide)	Vitavax 10% D, Vitavax 75% WP, Vitavax 34% liq. Vitaflow.	It is a systemic fungicide used for seed treatment of cereals against bunts and smuts, especially loose smut of wheat

<p>2. Oxycarboxin (5,6-dihydro-2-methyl-1,4-oxathin-3-carboxianilid-4,4-dioxide)</p>	<p>Plantvax5G, Plantvax5%1 iq. Plantvax1.5EC, 10% dust, 75 WP.</p>	<p>It is a systemic fungicide used for the treatment of rust diseases of cereals, pulses, ornamentals, vegetables and coffee.</p>
<p>3. Pyracarbolid (2-methyl-5,6-dihydro-4H-Pyran-3-carboxylic acid anilide).</p>	<p>Sicarol.</p>	<p>It controls rusts, smuts of many crops and <i>Rhizoctonia solani</i>, but is slightly more effective than carboxin.</p>

## II. Benzimidazoles

The chemicals of this group show a very broad spectrum activity against a variety of fungi. However, they are not effective against bacteria as well as fungi belonging to *Mastigomycotina*.

Two types of fungicidal derivatives of benzimidazoles are known. The first type of derivatives includes fungicides such as thiabendazole and fuberidazole. The fungicidal moiety of the second type is methyl-2-benzimidazole carbamate (MBC). The fungicides of this group may be simple MBC such as carbendazim or a complex form such as benomyl, which transforms into MBC in plant system. Some of the important diseases controlled by these compounds are shown below:

Common Name	Trade Name	Diseases Controlled
<p>1. Benomyl (Methyl-10 (butyl carbomyl)-2 benzimidazole carbamate)</p>	<p>Benlate 50 WP, Benomyl, Bavistin 50 WP, MBC, Dersol, B. Sten 50, Zoom, Tagstin, Agrozim,</p>	<p>It is a protective and eradicated fungicide with systemic activity, effective against a wide range of fungi.</p>

<p>2. Carbendazim (MBC)(Methyl -2-benzimidazole carbamate)</p>	<p>Jkenstin.</p>	<p>affecting field crops, fruits and ornamentals. It is very effective against rice blast, apple scab, powdery mildew of cereals, rose, cucurbits and apple and Diseases caused by <i>Verticillium</i> and <i>Rhizoctonia</i>. It is also used as pre- and postharvest sprays of dips for the control of storage rot of fruits and vegetables. Carbendazim is a systemic fungicide controlling a wider range of fungal pathogens of field crops, fruits, ornamentals and vegetables. It is used as spray, seedling dip, seed treatment, soil dressing and as post harvest treatment of fruits. It is very effective against wilt diseases especially, banana wilt. It controls effectively the Sigatoka leaf spot of banana, turmeric leaf spot and rust diseases in many crops.</p>
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3. Thiabendazole (TBZ) (2,4-thiazoylbenzimidazole)	Thiabendazole, Mertect, Tecto, Storite.	It is a broad spectrum systemic fungicide effective against many major fungal diseases. Pathogenic fungal control
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<p>4. Fuberidazole (2, (2-buryl)-benzimidazole).</p>	<p>Voronit.</p>	<p>includes species of <i>Botrytis</i>, <i>Ceratocystis</i>, <i>Cercospora</i>, <i>Colletotrichum</i>, <i>Fusarium</i>, <i>Rhizoctonia</i>, <i>Sclerotinia</i>, <i>Septoria</i> and <i>Verticillium</i>. It is also effective for the post harvest treatment of fruits and vegetables to control storage diseases.</p> <p>It is used for the treatment of seeds against diseases caused by <i>Fusarium</i>, particularly <i>F. nivale</i> on rye and <i>F. culmorum</i> of peas</p>
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### III. Thiophanates

These compounds represent a new group of systemic fungicides based on thiourea. They are the derivatives of thioallophanic acid. These fungicides contain aromatic nucleus which is converted into benzimidazole ring for their activity. Hence, thiophanates are often classified under benzimidazole group and the biological activity of thiophanates resembles of benomyl. Two compounds are developed under this group are discussed.

Common Name	Trade Name	Diseases Controlled
<p>1. Thiophanate (1,2-bis (ethyl carbonyl-2-thioureido)benzene)</p>	<p>Topsin 50 WP, Cercobin 50 WP, Enovit.</p>	<p>It is a systemic fungicide with a broad range of action, effective against</p>

<p>2. Thiophanate-methyl (1,2-bis(3-methoxycarbonyl-2-thioureido)benzene.)</p>	<p>Topsin-M70 WP, Cercobin-M 70 WP, Envovit-methyl, Mildothane.</p>	<p><i>Venturia</i> spp., on apple and pear crops, powdery mildews, <i>Botrytis</i> and <i>Sclerotinia</i> spp. On various crops.  It is effective against a wider range of fungal pathogens, including <i>Venturia</i> spp. on apples and pears, <i>Mycosphaerella musicola</i> on bananas, powdery mildew on apples, cucurbits, pears and vines, <i>Pyricularia oryzae</i> on rice, <i>Botrytis</i> and <i>Cercospora</i> on various crops.</p>
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#### IV. Morpholines

Common Name	Trade Name	Diseases Controlled
<p>Tridemorph (2,6-dimethyl-4-cyclo - tridecyl morpholine)</p>	<p>Calixin 75EC, Bardew, Beacon</p>	<p>It is a radical fungicide with systemic action, being absorbed through foliage and roots to give some protective action. It controls powdery mildew diseases of</p>

		cereals, vegetables and ornamentals. It is highly effective against <i>Mycosphaerella</i> , <i>Exobasidium</i>
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**V. Pyrimidines, Pyridines, Piperidines and Imidazole**

Common Name	Trade Name	Diseases Controlled
1. Triadimefon (1-(4-chlorophenoxy)-3,3-dimethyl-1-(1,2,4-triazol-1-yl)butan-2-one)	Bayleton, Amiral	It is very effective against powdery mildews and rusts of several crops.
2. Triadimenol (1-(4-Chlorophenoxy)-3,3-dimethyl-1-(1,2,4-triazol-1-yl)butan-2-ol)	Baytan	It is also very effective against powdery mildews and rusts of several crops.
3. Bitertanal (B-(1-(1-biphenyl-4-yloxy)-1,2,4-triazole-1-ethyl)-1-H-1,2,4-triazole-1-ethanol)	Baycor  Terrazole 30% WP, Terrazole 95% WP,	It is highly effective against rusts and powdery mildew of a variety of crops. It is also used against <i>Venturia</i> and <i>Monilinia</i> on fruits and <i>Cereospora</i> leaf spots of groundnut and banana.

<p>4. Etridiazole (5-ethoxy-3-trichloromethyl,1,2-4-thiadiazole)</p>	<p>Terrazole 25% EC, Koban, Pansol EG, Pansol 4% DP, Turban WP, Terracoat Aaterra.</p>	<p>It is very effective against <i>Phytophthora</i> and <i>Pythium</i> spp. and seeding diseases of cotton, groundnut, vegetables, fruits and ornamentals</p>
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**VI. Hydroxy Pyrimidines**

Common Name	Trade Name	Diseases Controlled
<p>1. Ethirimol (5-butyl 2-ethylamino-4-hydroxy-6-methylpyrimidine)</p>	<p>Milliaterm 80 WDP, Milcurb Super, Milgo</p>	<p>It is effective against powdery mildew of cereals and other field crops. It is also effective against powdery mildews of cucumber and ornamentals.</p>
<p>2. Dimethirimol (5-butyl 2-dimethylamino-4-hydroxy-6-methylpyrimidine)</p>	<p>Milcurb</p>	<p>It is very effective against powdery mildews of chrysanthemum and cucurbits.</p>
<p><b>VII. Furanderivatives</b></p>		
<p>1. Furcarbanil (2-5-dimethyl-3-furanilide)</p>		<p>It is used as seed or soil application, it systemically controlled bean rust and is being used as a seed</p>

<p>2. Cyclafuramid(N-cyclohexyl-2,5-dimethylfiramide)</p> <p><b>VIII. Benzanilide derivative</b> 1. Mebenil (2-methylbenzanilide)</p>		<p>dressing fungicide against loosesmut of wheat and barley.</p> <p>It is effective against bunts, smuts and rusts of cereals, coffee rust, blister blight of tea, smut and red rot of sugarcane, <i>Fusarium wilt</i> of tomato, <i>Rhizoctonia</i> on tomato, potato, groundnut, rice as well as <i>Armillaria</i> sp. On rubber.</p> <p>It is effective against yellow rust on wheat and barley (<i>P. striiformis</i>) and brown rust on barley (<i>P. hordei</i>). It is also having direct fungitoxic activity against <i>Sclerotium rolfsii</i> and <i>Rhizoctonia</i>.</p>
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**IX. Organophosphorous compounds**

Common Name	Trade Name	Diseases Controlled
1. Pyrazophos (2-0-0-	Afugan, Curamil, WP,	It is used to control

<p>Diethylthionophosphoryl)-5-methyl-6-carbethoxy-pyrazolo-(1-5a)pyrimidine)</p>	<p>Missile EC.</p>	<p>powdery mildew of cereals, vegetables, fruits and ornamentals.</p>
<p>2. Iprobenphos (IBP) (S-benzyl-0-0-bis(isopropylphosphorothiate)</p>	<p>Kitazin 48% EC, Kitazin 17G, Kitazin 2% D.</p>	<p>It is used to control <i>Pyricularia oryzae</i> and sheath blight of rice.</p>
<p><b>X. Piperazine</b> 1. Triforine (N,N-bis-(1-formamido-2,2,2-trichloroethyl)-piperazine)</p>	<p>Saprol-EG, Fungitex.</p>	<p>It is effective against powdery mildew, scab and other diseases of fruits and rust on ornamentals and cereals.</p>
<p><b>XI. Phenol derivative</b> 1. Chloroneb (1-4-dichloro-2,5-dimethoxybenzene)</p>	<p>Demonsan 65 WP, Tersan SP, Turf Fungicide</p>	<p>It is also active against storage diseases of fruits. It is highly fungistatic to <i>Rhizoctonia</i> spp., moderately so to <i>Pythium</i> spp. and poorly to <i>Fusarium</i> spp. It is used as a supplemental seed treatment for beans and soybeans to control seedling disease.</p>

**XIII. Other systemic fungicides**

Common Name	Trade Name	Diseases Controlled
1. Metalaxyl (methyl-DLN-(2,6-dimethylphenyl-N)-2-methoxyacetyl	Apron 35 SD, Ridomil	It is a systemic fungicide and highly effective for specific use as seed dressing against fungal pathogens of the order Peronosporales.
2. Metalaxyl + Mancozeb	Ridomil MZ 72 WP (8% Metalaxyl + 64% Mancozeb) Beam, Bim Alliette 80 WP	It is a fungicide with systemic and contact action and effective against damping-off, root rots, stem rots, and downy mildew of grapes and millets.  It is a specific fungicide used against paddy blast fungus, <i>P. oryzae</i>
3. Tricyclazole (5-methyl-1,2,4-triazole(3,4b)-benzothiazole)		It is a very specific fungicide for Oomycetous fungi, especially against <i>Pythium</i> and <i>Phytophthora</i>
4. Fosetyl AI (Aluminium-Trisaluminium		

**Antibiotics**

Antibiotic is defined as a chemical substance produced by a micro-organism which in low concentration can inhibit or even kill other micro-

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organism. Because of their specificity of



action against plant pathogens, relatively low phytotoxicity, absorption through foliage and systemic translocation and activity in low concentration, the use of antibiotic is becoming very popular and very effectively used in managing several plant diseases. They can be grouped as antibacterial antibiotics and antifungal antibiotics. Most antibiotics are products of several actinomycetes and a few are from fungi and bacteria.

## **I. Antibacterial antibiotics**

### **1. Streptomycin sulphate**

Streptomycin is an antibacterial antibiotic produced by *Streptomyces griseus*. Streptomycin as streptomycin sulphate is sold as Agrimycin-100, Streptomycin sulphate, Plantomycin, Streptocycline, Paushamycin, Phytostrip, Agristrep and Embamycin. Agrimycin-100 contains 15 per cent streptomycin sulphate + 1.5 percent terramycin (Oxy tetracycline). Agristrep contains 37 per cent streptomycin sulphate.

Phytopomycin contains 20 per cent streptomycin. Streptocycline and paushamycin contains 9 parts of streptomycin and 1 part of tetracycline hydrochloride.

This group of antibiotics act against a broad range of bacterial pathogens causing blights, wilt, rots etc. This antibiotic is used at concentrations of 100-500 ppm. Some important diseases controlled are blight of apple and pear (*Erwinia amylovora*), Citrus canker (*Xanthomonas campestris p.v. citri*), Cotton black arm (*X.c. p.v. malvacearum*), bacterial leaf spot of tomato (*Pseudomonas solanacearum*), wild fire of tobacco (*Pseudomonas tabaci*) and soft rot of vegetables (*Erwinia carotovora*).

In addition, it is used as a dip for potato seed pieces against various bacterial rots and as a disinfectant in bacterial pathogens of beans, cotton, crucifers, cereals and vegetables. Although it is an antibacterial antibiotic, it is also effective against some diseases caused by Oomycetous fungi, especially foot-rot and leaf-rot of betel vine caused by *Phytophthora parasitica* var. *piperina*.

### **2. Tetracyclines**

Antibiotics belonging to this group are produced by many species of *Streptomyces*. This group includes Terramycin or Oxymycin (Oxytetracycline). All these antibiotics are bacteriostatic, bactericidal and mycoplasma static. These are very effective against seed-borne bacteria. This group of antibiotic is very effective in managing MLO diseases of a wide range of crops. These are mostly used as combination products with Streptomycin sulphate in controlling

a wider range of bacterial diseases. Oxytetracyclines are effectively used as soil drench or as root dip control for crown gall diseases in rosaceous plants caused by *Agrobacterium tumefaciens*.

## **II Antifungal antibiotics**

### **1. Aureofungin**

It is a heptaene antibiotic produced in sub-merged culture of *Streptomyces verticillium cinnamomeum* var. *terricola*. It is absorbed and translocated to other parts of the plants when applied as spray or given to roots as drench. It is sold as Aureofungin-Sol. Containing 33.3% Aureofungin and normally sprays at 50-100 ppm. The diseases controlled are citrus gummosis caused by several species of *Phytophthora*, powdery mildew of apple caused by *Podosphaera leucotricha* and apple scab (*Venturia inaequalis*), groundnut tikka leaf spot, downy mildew, powdery mildew and anthracnose of grapes, potato early and late blight. As seed treatment it effectively checked are *Diplodia* rot of mango, *Alternaria* rot of tomato, *Pythium* rot of cucurbits and *Penicillium* rot of apples and citrus. As a truck application/root feeding, 2 g of aureofungin-sol + 1 g of copper sulphate in 100 ml of water effectively reduce Thanjavur wilt of coconut.

### **2. Griseofulvin**

This antifungal antibiotic was first discovered to be produced by *Penicillium griseofulvum* and now by several species of *Penicillium*, viz., *P. patulum*, *P. nigricans*, *P. urticae*, and *P. raciborskii*. It is commercially available as Griseofulvin, Fulvicin and Grisovin. It is highly toxic to powdery mildew of beans and roses, downy mildew of cucumber. It is also used to control *Alternaria solani* in tomato, *Sclerotinia fructigena* in apple and *Botrytis cinerea* in lettuce.

### **3. Cycloheximide**

It is obtained as a by-product in streptomycin manufacture. It is produced by different species of *Streptomyces*, including *S. griseus* and *S. nouresi*. It is commercially available as Actidione, Actidione PM, Actidione RZ and Actispray. It is active against a wide range of fungi and yeast. Its use is limited because it is extremely phytotoxic. It is effective against powdery mildew of beans (*Erysiphe polygoni*), Bunt of wheat (*Tilletia* spp.) brown rot of peach (*Sclerotinia fructicola*) and postharvest rots of fruits caused by *Rhizopus* and *Botrytis* spp.

### **4. Blasticidin**

It is a product of *Streptomyces griseochromogenes* and specifically used against blast disease of rice caused by *Pyricularia oryzae*. It is commercially sold as Bla-s.

### **5. Antimycin**

It is produced by several species of *Streptomyces*, especially *S. griseus* and *S. Kitasawensis*. It is effectively used against early blight of tomato, rice blast and seedling blight of oats. It is commercially sold as Antimycin.

### **6. Kasugamycin**

It is obtained from *Streptomyces kasugaensis*. It is also very specific antibiotic against rice blast disease. It is commercially available as Kasumin.

### **7. Thiolutin**

It is produced by *Streptomyces albus* and effectively used to control late blight of potato and downy mildew of cruciferous vegetables.

### **8. Endomycin**

It is a product of *Streptomyces endus* and effectively used against leaf rust of wheat and fruit rot of strawberry (*Botrytis cinerea*).

### **9. Bulbiformin**

It is produced by a bacterium, *Bacillus subtilis* and is very effectively used against wilt diseases, particularly redgram wilt.

### **10. Nystatin**

It is also produced by *Streptomyces noursei*. It is successfully used against anthracnose disease of banana and beans. It also checks downy mildew of cucurbits. As a post harvest dip, it effectively reduces brown rot of peach and anthracnose of banana in storage rooms. It is commercially marketed as Mycostain and Fungicidin.

### **11. Eurocidin**

It is a pentaene antibiotic produced by *Streptomyces anandii* and called as pentaene G-8. It is effectively used against diseases caused by several species of *Colletotrichum* and *Helminthosporium*.

### **Methods of application of fungicides – Precautions and safety measures while handling fungicides**

Proper selection of a fungicide and its application at the correct dose and the proper time are highly essential for the management of plant diseases. The basic requirement of an application method is that it delivers the fungicide to the site where the active compound will

prevent the fungus damaging the plant. This is mostly achieved by spray, fog, smoke, aerosol, mist, dust, or granules applied to the growing plant or by seed or soil treatment.

In addition, some trees and shrubs can be protected by injection of fungicide liquid into the trunk or by brushing wounds with fungicide paints or slurries. In the case of sprays, mists, aerosols and fogs, the fungicide is in the form of droplets of water or another fluid. In the case of smokes, the solid particles of the fungicide are carried by the air. In the case of dusts and granules, the fungicide is straightly mixed with an inert carrier, impregnated into it coated on the particles, which are applied mechanically.

The object of spraying or dusting is to cover the entire susceptible surface of host with a thin covering of a suitable concentration of the fungicide before the pathogen has come into contact with the host. However, these practices may not effectively eradicate the inoculum present on the surface of the seeds or deep-seated in the seed. So, the application of chemicals as seed dressing is highly essential.

In addition, soil harbours several pathogens which cause root diseases in several crop plants. So treatment of soil with chemicals is also highly useful in reducing the inoculum load present in the soil. The fungicidal application varies according to the nature of the host, the disease and nature of survival and spread of the pathogen. The methods which are commonly adopted in the application of the fungicides are discussed.

### **1. Seed treatment**

The seed treatment with fungicides is highly essential because a large number of fungal pathogens are carried on or in the seed. In addition, when these seeds are sown, they are also vulnerable to attack by many common soil-borne pathogens, leading to either seed rot, seedling mortality or produce diseases at a later stage. Seed treatment is probably the most effective and economic method of disease control and is being advocated as a regular practice in crop protection against soil and seed-borne pathogens. Seed treatment is therapeutic when it kills pathogens that infect embryos, cotyledons or endosperms under the seed coat, eradicates them when they kill pathogens that contaminate seed surfaces and protective when it prevents penetration of soil-borne pathogens into the seedling. There are various types of seed treatment and broadly they may be divided into three categories (a) Mechanical, (b) Chemical and (c) Physical.

### **A. Mechanical method**

Some pathogen when attack the seeds, there may be alteration in size, shape and weight of seeds by which it is possible to detect the infected seeds and separate them from the healthy ones. In the case of ergot diseases of cumbu, rye and sorghum, the fungal sclerotia are usually larger in size and lighter than healthy grains. So by sieving or flotation, the infected grains may be easily separated. Such mechanical separation eliminates the infected grains may be easily separated. Such mechanical separation eliminates the infected materials to a larger extent. This method is also highly useful to separate infected grains in the case of „tundu“ disease of wheat. Eg. Removal of ergot in cumbu seeds.

Dissolve 2kg of common salt in 10 litres of water (20% solution). Drop the seeds into the salt solution and stir well. Remove the ergot affected seeds and sclerotia which float on the surface. Wash the seeds in fresh water 2 or 3 times to remove the salts on the seeds. Dry these seeds in shade and use for sowing.

### **B. Chemical methods**

Using fungicides on seed is one of the most efficient and economical methods of chemical disease control. On the basis of their tenacity and action, the seed dressing chemicals may be grouped as (i) Seed disinfectant, which disinfect the seed but may not remain active for a long period after the seed has been sown and (ii) Seed protectants, which disinfect the seed surface and stick to the seed surface for some time after the seed has been sown, thus giving temporary protection to the young seedlings against soilborne fungi. Now, the systemic fungicides are impregnated into the seeds to eliminate the deep seated infection in the seeds. These seed dressing chemicals may be applied by (i) Dry treatment (ii) Wet treatment and (iii) Slurry.

#### **(i) Dry Seed Treatment**

In this method, the fungicide adheres in a fine film on the surface of the seeds. A calculated quantity of fungicide is applied and mixed with seed using machinery specially designed for the purpose. The fungicides may be treated with the seeds of small lots using simple Rotary seed Dresser (Seed treating drum) or of large seed lots at seed processing plants using Grain treating machines. Normally in field level, dry seed treatment is carried out in dry rotary seed treating drums which ensure proper coating of the chemical on the surface of seeds. In addition, the dry dressing method is also used in pulses, cotton and oil seeds with the

antagonistic fungus like *Trichoderma vitide* by mixing the formulation at the rate of 4g/kg of the seed.

Eg. Dry seed treatment in paddy.

Mix a required amount of fungicide with required quantity of seeds in a seed treating drum or polythene lined gunny bags, so as to provide uniform coating of the fungicide over these seeds. Treat the seeds at least 24 hours prior to soaking for sprouting. Any one of the following chemical may be used for treatment at the rate of 2g/kg : Thiram or Captan or Carboxin or Tricyclazole.

### **(ii) Wet seed treatment**

This method involves preparing fungicide suspension in water, often at field rates and then dipping the seeds or seedlings or propagative materials for a specified time. The seeds cannot be stored and the treatment has to be done before sowing. This treatment is usually applied for treating vegetatively propagative materials like cuttings, tubers, corms, setts, rhizomes, bulbs etc., which are not amenable to dry or slurry treatment.

#### **a. Seed dip / Seed soaking**

For certain crops, seed soaking is essential. Seeds treated by these methods have to be properly dried after treatment. The fungicide adheres as a thin film over the seed surface which gives protection against invasion by soil-borne pathogens.

Eg. Seed dip treatment in paddy.

Prepare the fungicidal solution by mixing any of the fungicides viz., carbendazim or pyroquilon or tricyclazole at the rate of 2g/litre of water and soak the seeds in the solution for 2 hrs. Drain the solution and keep the seeds for sprouting.

Eg. Seed dip treatment in Wheat.

Prepare 0.2% of carboxin (2g/litre of water) and soak the seeds for 6 hours. Drain the solution and dry the seeds properly before sowing. This effectively eliminates the loose smut pathogen, *Ustilago nuda tritici*.

#### **b. Seedling dip / root dip**

The seedlings of vegetables and fruits are normally dipped in 0.25% copper oxychloride or 0.1% carbendazim solution for 5 minutes to protect against seedling blight and rots.

**c. Rhizomedip**

The rhizomes of cardamom, ginger and turmeric are treated with 0.1% emisan solution for 20 minutes to eliminate root causing pathogen present in the soil.

**d. Settdip /Suckerdip**

The sets of sugarcane and tapioca are dipped in 0.1% emisan solution for 30 minutes. The suckers of pineapple may also be treated by this method to protect from soilborne diseases.

**(iii) Slurry treatment (Seed pelleting)**

In this method, chemical is applied in the form of a thin paste (active material is dissolved in small quantity of water). The required quantity of the fungicide slurry is mixed with the specified quantity of the seed so that during the process of treatment slurry gets deposited on the surface of seeds in the form of a thin paste which later dries up.

Almost all the seed processing units have slurry treaters. In these, slurry treaters, the requisite quantity of fungicide slurry is mixed with specified quantity of seed before the seed lot is bagged. The slurry treatment is more efficient than the rotary seed dressers.

Eg. Seed pelleting in ragi.

Mix 2.5g of carbendazim in 40 ml of water and add 0.5g of gum to the fungicidal solution. Add 2kg of seed to this solution and mix thoroughly to ensure a uniform coating of the fungicide over the seed. Dry the seeds under the shade. Treat the seeds 24 hrs prior to sowing.

**(iv) Special method of seed treatment**

Eg. Acid -delinting in cotton

This is followed in cotton to kill the seed-borne fungi and bacteria. The seeds are treated with concentrated sulphuric acid @ 100 ml/kg of seed for 2-3 minutes. The seeds are then washed 2 or 3 times thoroughly with cold water and shade dried. After drying, they are again treated with captan or thiram @ 4g/kg before sowing.

**II. Soil treatment**

It is well known that soil harbours a large number of plant pathogens and the primary sources of many plant pathogens happens to be in soil where dead organic matter supports active or dormant stages of pathogens. In addition, seed treatment does not afford sufficient

protection against seedling diseases and a treatment of soil around the seed is necessary to protect them.

Soil treatment is largely curative in nature as it mainly aims at killing the pathogens in soil and making the soil safe for the growth of the plant.

Chemical treatments of the soil is comparatively simple, especially when the soil is fallow as the chemical is volatile and disappears quickly either by volatilization or decomposition. Soil treating chemicals should be non-injurious to the plants in the soil adjacent to the area where treatment has been carried out because there may be standing crop in adjacent fields. The soil treatment methods involving the use of chemicals are

(i) Soil drenching, (ii) broadcasting, (iii) furrow application, (iv) fumigation and (v) chemigation.

**(i) Soil drenching**

This method is followed for controlling damping off and root rot infections at the ground level. Requisite quantity of fungicide suspension is applied per unit area so that the fungicide reaches to a depth of at least 10-15 cm.

Eg. Emisan, PCNB, Carbendazim, Copper fungicides, etc.

**(ii) Broadcasting**

It is followed in granular fungicides where the pellets are broadcasted near the plant.

**(iii) Furrow application**

It is done specifically in the control of some diseases where the direct application of the fungicides on the plant surface results in phytotoxicity. It is specifically practiced in the control of powdery mildew of tobacco where the sulphur dust is applied in the furrows.

**(iv) Fumigation**

Volatile toxicants (fumigants) such as methyl bromide, chloropicrin, formaldehyde and vapam are the best chemical sterilants for soil to kill fungi and nematodes as they penetrate the soil efficiently. Fumigations are normally done in nursery areas and in glasshouses. The fumigant is applied to the soil and covered by thin polythene sheets for 5-7 days and removed. For example, formaldehyde is applied at 400 ml/100 Sq.m. The treated soil was irrigated and used 1 or 2 weeks later. Vapam is normally sprinkled on the soil surface and covered. Volatile liquid fumigants are also injected to a depth of 15-20 cm, using sub-soil injectors.

**(v) Chemigation**

In this method, the fungicides are directly mixed in the irrigation water. It is normally adopted using sprinkler or drip irrigation system.



### **III. Foliar application**

#### **A. Spraying**

This is the most commonly followed method. Spraying of fungicides is done on leaves, stems and fruits. Wettable powders are most commonly used for preparing spray solutions. The most common diluent or carrier is water. The dispersion of the spray is usually achieved by its passage under pressure through nozzle of the sprayer.

The amount of spray solution required for a hectare will depend on the nature of crops to be treated. For trees and shrubs more amount of spray solution is required than in the case of ground crops. Depending on the volume of fluid used for coverage, the sprays are categorised into high volume, medium volume, low volume, very high volume and ultra low volume.

The different equipments used for spray application are: Foot-operated sprayer, rock sprayer, knapsack sprayer, motorised knapsack sprayer (Power sprayer), tractor mounted sprayer, mist blower and aircraft or helicopter (aerial spray).

#### **B. Dusting**

Dusts are applied to all aerial parts of a plant as an alternative to spraying. Dry powders are used for covering host surface. Generally, dusting is practicable in calm weather and a better protective action is obtained if the dust is applied when the plant surface is wet with dew or raindrops. The equipments employed for the dusting operation are: Bellow duster, rotary duster, motorised knapsack duster and aircraft (aerial application).

### **IV. Post-harvest application**

Fruits and vegetables are largely damaged after harvest by fungi and bacteria. Many chemicals have been used as spray or dip or fumigation. Post-harvest fungicides are most frequently applied as aqueous suspensions or solutions. Dip application has the advantage of totally submerging the commodity so that maximum opportunity for penetration to the infection sites.

Systemic fungicides, particularly thiabendazole, benomyl, carbendazim, metalaxyl, fosetyl-AI have been found to be very effective against storage diseases. In addition, dithiocarbamates and antibiotics are also applied to control the post-harvest diseases. Wrapping the harvested products with fungicide impregnated wax paper is the latest method available.

## **VI. Special method of applications**

### **1. Trunk Application/Trunk Injection**

It is normally adopted in coconut garden to control Thanjavur wilt caused by *Ganoderma lucidum*. In the infected plant, a downward hole is made to a depth of 3-4" at an angle of 45° to the height of 3" from the ground level with the help of an auger. The solution containing 2g of Aureofungin soil and 1 g of copper sulphate in 100 ml of water is taken in a saline bottle and the bottle is tied with the tree. The hose is inserted into the hole and the stopper is adjusted to allow the solution in drops. After the treatment, the hole is covered with clay.

### **2. Root Feeding**

Root feeding is also adopted for the control of Thanjavur wilt of coconut instead of trunk application. The root region is exposed; actively growing young root is selected and given a slanting cut at the tip. The root is inserted into a polythene bag containing 100 ml of the fungicidal solution. The mouth of the bag is tied tightly with the root.

### **3. Pseudostem Injection**

This method is very effective in controlling the aphid vector (*Pentalonia nigronervosa*) of bunchy top of banana. The banana injector is used for injecting the insecticide. Banana injector is nothing but an Aspee baby sprayer of 500 ml capacity. In which, the nozzle is replaced by a lock system and aspirator needle No. 16. The tip of the needle is closed and two small holes are made in opposite direction.

It is for free flow of fluid and the lock system prevents the needle from dropping from the sprayer. One ml of monocrotophos mixed with water at 1:4 ratio is injected into the pseudostem of 3 months old crop and repeated twice at monthly intervals. The same injector can also be used to kill the bunchy top infected plants by injecting 2 ml of 2, 4-D (Femoxone) mixed in water at 1:8 ratio.

### **4. Corn Injection**

It is an effective method used to control Panama wilt of banana caused by *Fusarium oxysporum* f. sp. *cubense*. Capsule applicator is used for this purpose. It is nothing but an iron rod of 7 mm thickness to which a handle is attached at one end. The length of the rod is 45 cm and an iron plate is fixed at a distance of 7 cm from the tip.

The corm is exposed by removing the soil and a hole is made at a 45° angle to a depth of 5 cm. One or two gelatin capsules containing 50-60 mg of carbendazim is pushed in slowly and covered with soil. Instead of capsule, 3 ml of 2% carbendazim solution can also be injected into the hole.

### **5. Paring and Pralinage**

It is used to control *Fusarium* wilt and burrowing nematode (*Radopholus similis*) of banana. The roots as well as a small portion of corm is removed or chopped off with a sharp knife and the sucker is dipped in 0.1% carbendazim solution for 5 minutes.

Then, the sucker is dipped in clay slurry and furadan granules are sprinkled over the corm @ 40g/corm.

## Lecture 10

### **Host plant resistance – Importance – disease resistance, tolerance, susceptibility and disease escape. Horizontal and vertical resistance – Method of management of resistance.**

#### **Immunization – Systemic acquired resistance**

#### **Host plant resistance**

A physiological deviation from the normal functioning of the organism (i.e., the crop plant) caused by pathogenic organisms is a disease and may be caused by fungi, bacteria or viruses. The inherent ability of an organism (i.e., the crop plant) to resist or withstand the pathogen is called resistance. Disease resistance commonly met within the plant kingdom relative in nature, total immunity being too rare. Its hereditary transmission from parent to off-spring is essentially “Mendelian”, but often polygenic.

The earliest demonstration of the behaviour of “disease-resistance” as a character transmissible from parent to off-spring in the “Mendelian” fashion was given by Biffen (1905) in his work on yellow rust of wheat. Since then, intensive work has been done on this aspect which has proved the value of applying genetical principles in developing disease-resistant varieties of plants for effective control of diseases.

Resistant varieties can be the simplest, practical, effective and economical method of plant disease control. The use of resistant varieties cannot only ensure protection against diseases but also save the time, energy and money spent on other measures of control. In addition to these advantages, resistant varieties, if evolved, can be the only practical method of control of such diseases as viruses, phytoplasmas, wilts, and rusts etc. in which chemical control is very expensive and impractical.

In crops of low cash value, chemical and other methods of control are often too expensive to be applied. In such crops development of varieties resistant to important diseases can be an acceptable recommendation for the farmer. Pathogenicity is the ability of a pathogen to attack a host. Pathogenicity includes virulence and aggressiveness. Virulent strains of pathogen cause much severe symptoms of the disease and they carry the virulence gene that enables it to attack a particular host genotype.

Virulence is due to the action of one or a few genes. An aggressive strain of a pathogen causes severe disease on all the host genotypes which they are able to attack and aggressiveness is polygenically inherited. Host–

Pathogen relationship A disease is the result of an interaction

of genes governing resistance in the host with those governing pathogenicity in the pathogen. The resistance of a crop to a physiological race of the pathogen depends not only on the genotype of the host for resistance, but also upon the genotype of the pathogen for virulence or aggressiveness. Flor (1942) proposed the gene-for-gene hypothesis, according to which, for every gene for resistance in the host, there is a corresponding gene for pathogenicity in the pathogen.

It means that there are at least two alleles at a locus controlling resistance/susceptibility in the host (R-r) and two alleles at a corresponding locus in the pathogen (V-v) controlling virulence / aggressiveness. Out of the four possible interactions between these alleles, only one combination leads to the expression of resistance. The demonstration of gene-for-gene relationship requires genetic studies of both the host and the pathogen.

### **Pathogen**

VI v1 + Pathogen can infect; the host is R1 -+ susceptible r1 + + -  
Pathogen cannot infect; the host is resistant

The demonstration of gene-for-gene relationship requires genetic studies of both the host and the pathogen

### **Vertical resistance (VR) and horizontal resistance (HR)**

Van der Plank (1960) has discussed the whole issue of disease resistance in a different perspective. He calls the unstable and often complete type of resistance as vertical resistance and the more stable but somewhat incomplete resistance as horizontal resistance. If resistance to some races of a pathogen is more than to other races, it is called Vertical resistance. It is also called Perpendicular resistance, Physiological resistance, seedling resistance, hypersensitivity, race specific resistance or qualitative resistance. As it is conditioned by one or a few genes, it is called major gene or monogenic or oligogenic resistance.

Resistance to more than one race of the pathogen or to many or all races of the pathogen is called Horizontal Resistance. It is non-specific resistance governed by polygenes. It is severally termed as non-specific, general, polygenic, minor gene, mature plant, adult, quantitative resistance, partial or field resistance or tolerance. HR causes reduction in the number and rate of sporulation of the pathogen on the host and slows down the infection rate. HR includes tolerance, slow development of disease, escape and exclusion mechanisms besides

hypersensitive reaction. The difference between vertical resistance and horizontal resistance are given in table.

**Differences between vertical and horizontal disease resistance\* Detectable by analysis of variance on a suitable experiment**

<b>Feature</b>	<b>Vertical resistance</b>	<b>Horizontal resistance</b>
Pathotype-specificity	Race specific	Race non-specific
Nature of gene action	Oligogenic	Polygenic; rarely oligogenic
Response to pathogen	Usually, hypersensitive	Resistant response
Phenotypic expression	Qualitative	Quantitative
Stage of expression	Seedling to maturity	Expression increases as plant matures (Adult plant)
Selection and evaluation	Relatively easy	Relatively difficult
Risk of „boom and bust“	Present (rarely durable)	Absent (durable)
Suitable for: a. Host b. Pathogen	Annuals but not perennials Immobile pathogen, e.g., Soil pathogens, but formobile air-borne, Pathogens	Both annuals and perennials All pathogens
Need for specific deployment of resistant varieties	Critical for success with mobile pathogens	None
Need for other control measures	Likely	Much less likely
Host-pathogen interaction*	Present	Absent
Efficiency	Highly efficient against specific races	Variable, but operates against all races

Vertical resistance to specific races is generally governed by a single (monogenic) dominant gene or by a few dominant genes. Some of these genes may be multiple alleles as in leaf rust gene, Lr2 that confers resistance to *Puccinia recondita tritici*. In that locus, four genes designated as Lr2a, Lr2b, Lr2c and Lr2d are present and are tightly linked. Each of these genes confers resistance to a different spectrum of races and hence can be differentiated from one another. Such multiple alleles exist on Sr9 locus of wheat for *P. graminis tritici* and gene Pi-k in rice for resistance to *Pyricularia grisea*. The tight linkage between the multiple alleles permits an efficient transfer of all these genes in one attempt.

„Horizontal resistance“ (HR) reduces the rate of disease spread and is evenly spread against all races of the pathogen. The low terminal disease severity in HR is assumed to result from polygenic resistance. Morphological features such as size of stomata, stomatal density per unit area, hairiness, waxiness and several others influence the degree of resistance expressed. Partial resistance, dilatory resistance, lasting resistance are some other terms coined for denoting horizontal resistance.

The phenomenon of slow rusting manifested as lesser number of pustules per unit leaf area, smaller size of uredosori and increased latent period in some wheat cultivars is a typical example of this type of resistance. Although it is preferable to use varieties that have both vertical and horizontal resistance, most of the resistant varieties carry only one or few (2 or 3) major genes of vertical resistance. If varieties are resistant only to some of the races of pathogen and if the pathogen is airborne, then new races evolve easily, as happens with cereal rusts, the powdery mildew and *Phytophthora infestans*. Appearance of new races lead to breakdown of resistance of the popular, ruling genotype. As a result, varieties with vertical resistance need to be replaced at frequent intervals.

### **Boom and bust cycle**

In varietal improvement programmes, it is easy to incorporate the monogenic vertical resistance genes. But the success of exploiting the monogenic host resistance invariably does not last long. Whenever a single gene-based resistant variety is widely adopted, the impact would be the arrival of new matching pathotypes.

These pathotypes soon build up in population to create epidemics and eventually the variety is withdrawn. This phenomenon is generally called “boom and burst”. To avoid the implications of boom and burst phenomenon, use of durable host resistance is advocated in several crops. Durable resistance remains effective even though it may be widely grown over a long period of time, in an environment that favours the disease. For example, oat variety, Red Rust Proof is still resistant against crown rust even after a hundred years. Wheat varieties, Thatcher and Lee have withstood stem rust for 55 and 30 years, respectively. Cappelle Desprez expresses at adult stage, a moderate resistance to yellow rust and this has been maintained for the last 20 years.

Two of the genes like Lr34 for resistance of leaf rust and Sr2 for resistance to stem rust have been recognized for durability. Wheat cultivars such as HD2189, HP1102, DL153-2, DL803-3 and DL802-2, which possess Lr34 with other gene combinations have a good degree of resistance and have become popular with growers. So far, there is no precise way available to identify the genetic components that are associated with durable resistance. Nor does dissociation of genes for virulence totally explain the basis of varietal durability, though it is likely to be the most plausible reason. Boom and burst cycle—a characteristic of vertical resistance. Resistance to virus and virus vectors. Resistance to plant pathogenic viruses is generally polygenic in nature.

For example, the host pathogen reaction to the barley yellow dwarf virus (BYDV) is controlled by a detectable single gene. The discovery of Yd2 gene in Ethiopian barley further confirms that against some of the viral diseases, vertical resistance is very much functional. Antibiosis is the most common phenomenon where the host plant metabolites interfere with the normal life and growth of the insects following feeding activity.

Invariably, the adult body weight, fecundity and various facets of multiplication of the insects are adversely affected. The number of life cycles completed in a given period of time is also less. Therefore, in plants that exhibit antibiosis towards crop maturity, there is a marked reduction in the level of pest infestation (virus vector population) and host damage. Mechanism of disease resistance or Nature of disease resistance. Disease resistance is governed by several in-built mechanisms of the host, plants against infection by the pathogen. They are disease escape, disease endurance or tolerance and true resistance.



**a. Disease escape**

It is a prevention mechanism that causes the host to escape pathogenic infection. Early or late maturity of the crop may prevent physical contact of the pathogen with the host. Mechanical and anatomical barriers such as thick cuticle, waxy bloom on leaves and stem, stomatal regulation prevent penetration of spores. Ergot, a fungal disease of inflorescence in cereals caused by *Claviceps purpurea* does not affect varieties of wheat and barley in which the flowers remain closed until pollination occurs. Erect leaves of barley avoid deposition of spores of *Erysiphe graminis tritici* in contrast to prostrate leaves. Early maturing varieties of groundnut escape early leaf spot infection (*Cercospora arachidicola*) and early varieties of wheat escape rust and loose smut infection.

A change in planting season has also been successfully employed as a measure of securing escape, e.g., the leaf rust of sugarcane (*Puccinia sacchari*) in the canal areas of Bombay severely affects cane when planted in June, but is of minor importance or absent in crops sown in October. Disease escape confers pseudo-resistance.

**b. Disease endurance**

The host after being infected by the pathogen tolerates the infection and suffers less damage. It does not result in any substantial decrease in yield. This is brought about by influence of external factors. It is a well-known phenomenon that plants fertilized with phosphatic and potash manures are more tolerant to disease; this is the case in wheat against rust infection. Rice crops fertilized by silicates are “resistant” to blast (*Pyricularia oryzae*) in Japan. Wheat crops fertilized by potash and phosphatic manures are highly tolerant to mildew and rust infection. The fertilizers act indirectly to arrest vegetative growth and promote early maturity, better straw and strengthening tissues to protect the plant which forms a bulwark against pathogenic invasion.

**c. True resistance**

It is the ability of the host plant to resist or withstand the attack of a pathogen. True resistance is inheritable and much less subject to environmental influence. It is specific in character. The basis of resistance may be morphological, functional, structural or protoplasmic. Functional nature of resistance is determined by opening of the stomata, time of opening of flowers and time of maturity, rate of cork formation and cambial activity.

Structural characters include the proportion of strengthening tissues, fibre content, nature of middle lamella, corky layers, number and structure of stomata and lenticels and their sizes. Protoplasmic factors controlling resistance are related to cell contents and include acids, tannins, anthocyanins, chemical constituents and their proportion, antibiotic activity and hypersensitivity present in the plant cells and in addition biological antagonism of the protoplasm of the host and the pathogen. True resistance, however, is of a specific character and is determined by the defence equipment and activities of the plant itself against the parasitic invasion and is therefore not subject to any appreciable modifications by external factors.

### **Methods of breeding for disease resistance**

The methods of breeding varieties resistant to diseases do not differ greatly from those adopted for other characters. The following methods are used:

1. Introduction,
2. Selection,
3. Hybridization followed by selection,
4. Backcross method,
5. Induced mutagenesis,
6. Development of multilines and
7. Tissue culture techniques

#### **1. Introduction**

It is a very simple and inexpensive method. Varieties resistant to a particular disease elsewhere may be thoroughly tested in the regions in which they are proposed to be introduced. Their yield performance and disease resistance should be confirmed by large scale cultivation. It is possible that a variety resistant in one region need not be resistant in another region due to variation in the physiological race of the pathogen or due to a much different agroclimatic condition in the new location.

Introductions have served as a useful method of disease control. For example, Ridley wheat introduced from Australia has been useful as a rust resistant variety. Manila, a rice variety introduced in Karnataka from the Philippines, has tolerance to blast, bacterial leaf blight and sheath blight. Intan, a Javanica type rice variety introduced in Karnataka from Indonesia is highly resistant to blast. Munal, a rice variety introduced in West Bengal from the U.S.A. is tolerant to blast, bacterial leaf blight and leaf folder (pest). Some of IRR rice varieties such as

IR 20, IR.24, IR.28, IR.34, IR.36 and IR .50 possess resistance to one or more diseases. Early varieties of groundnut introduced from U.S.A. have been resistant to leaf spot (*Cercosora arachidicola*).

Kalyan Sona and Sonalika wheat varieties originated from the segregating materials introduced from CIMMYT, Mexico and were rust resistant. Introductions also serve as sources

of resistance in breeding programmes. For example, African pearl millet (*P. americanum*) introductions have been used for developing downy mildew resistant male sterile lines (Tift 23A cytoplasm) for use in hybrid pearl millet production. This is an important development in the hybrid pearl millet programmes since the original male sterile lines Tift 23A and 23D2A were extremely susceptible to downy mildew. The introduction of Co.475 variety of sugarcane in Mumbai has conquered red rot but brought in leaf rust and whiplash to the fore.

## **2. Selection**

This is a better method than introduction and has more chances of success in obtaining disease-resistant plants. The work of selection is carried out either in the naturally infected

fields under field conditions or under artificially inoculated conditions. The resistance in such individuals will occur in nature by mutation. To ensure the resistant character of a plant, a large population of crop plants may be exposed to the attack of pathogen under artificial conditions and the non-infected plants may be chosen. Suvarnamodan rice of Kerala is a pure line of ARC.11775 and is highly tolerant to blast.

Sugandh of Bihar is a selection from Basmati rice of Orissa tolerant to bacterial leaf blight. Rice varieties Sudha (Bihar), Sabita, Nalini (West Bengal), Patel 85 (Madhya Pradesh), Janaki (Bihar), Improved White Ponni (Tamil Nadu), Ambika (Maharashtra), are some of the rice selections resistant to one or more diseases. MCU 1 cotton, a selection from Co 4, is resistant to Kufri Red, a potato selection from Darjeeling Red Round is a disease resistant variety.

## **3. Hybridization**

When selection of resistant varieties is not feasible, resistant varieties may be evolved by crossing the susceptible popular variety with resistant wild variety where in the resistant gene or genes transferred into the genetic make up of susceptible variety. Very often the F1 from crosses may be resistant but carry the other undesirable qualities of the resistant parent. The bad qualities are removed by several back crossings of F1 with the susceptible parent may ultimately

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yield resistant progeny with good agronomic characteristics.

Under certain circumstances pedigree or bulk method of selection is followed to obtain a resistant variety. In this method, the crosses are made till F<sub>2</sub> population is got. Selections are made in F<sub>2</sub> generation for superior genetic traits including disease resistance. By continued selfing, selections are made through F<sub>3</sub> to F<sub>5</sub> or F<sub>6</sub> generations and the best variety is selected. This method is suited for small grains and beans but unsuited to fruits and vegetables.

#### **4. Backcross method**

Back cross method is widely used to transfer disease resistance from wild species. Wild species are rice sources of disease resistance. Interspecific hybridization is made to transfer the gene or genes for resistance to the cultivated species. Resistance to grassy stunt virus from *Oryza nivara* to *O. sativa*, late blight resistance from *Solanum demissum* to cultivated potato, rust resistance from *durum* to *aestivum* wheat are some of the examples involving interspecific hybridization. Depending upon the number of genes governing resistance and the nature of the gene, whether dominant or recessive, the procedure varies. The number of back crosses to the cultivated species may be five to six. Once the backcross progeny resembles the cultivated parent, then they are selfed and segregating progeny screened for disease resistance.

#### **5. Induced mutagenesis**

While following mutation breeding for disease resistance, a large number of mutation progeny should be produced and screened under artificial epiphytotic condition to select resistant plants. MCU10 cotton, a resistant variety to bacterial blight was evolved in Tamil Nadu by subjecting seeds of a susceptible variety CO4 to gamma rays followed by rigorous screening and selection. Development of multilines The concept of multilines was first suggested by Jensen (1952) and developed by Borlaug (1959) for evolving multiline varieties to resist stem rust in wheat. A multiline variety is a composite of genetically similar lines, except that each line possesses a different gene for resistance to the pathogen.

Lines that are genetically similar, except for one gene, are called isolines. It is assumed that gene for resistance in each isolate contributes resistance to a separate physiological race or group of races. Genes for disease resistance are transferred by backcrossing from donor varieties to a common disease susceptible, but agronomically superior, recurrent parent. Isolines are generated differing only in the gene for disease resistance. The isolines are composited to synthesize a multiline variety. The isolines are maintained for synthesizing the multiline

whenever needed. A multiline variety is composed of a mixture of resistant and susceptible genotypes and provides a buffering effect against rapid development of disease. It will provide resistance or tolerance to a broad spectrum of races of a pathogen. If new races of the pathogen are identified at a later stage, additional isolines resistant to the newly arisen races may be constituted and incorporated.

Care should be taken to see that there is uniformity for height, maturity and other features in the multiline. Though multilines provide stability of yield due to reduction of damage by pathogens, the limitations of multiline varieties are that the yield level of multiline varieties is limited to that of the recurrent parent, 4 to 5 years are required to stabilize isogenic lines and the pathogen may produce new races at a faster rate than the development of a multiline. Multiline varieties have been developed for resistance to stem rust and stripe rust of wheat and crown rust of oats. The first multiline variety in wheat, „Miramar 60“ was developed and released in Columbia to combat the attack of yellow rust. „Miramar 63“ and „Miramar 65“ were resistant to stem rust and stripe rust. „Yoqui 50“, „Crew“ and „Tumult“ are a few other wheat multilines. Kalyan sona and Sonalika-based multilines of wheat resistant to different races of rust have been developed in India.

### **7. Tissue culture technique**

Tissue culture techniques to produce somaclonal variation for disease are developed in different crops. Somaclonal variations for disease resistance are reported in *Zeamays* for *Drechslera maydis* race T-toxin resistance, in *Brassica napus* for resistance/tolerance to *Phoma lingam*, early and late blight resistance in potato, *Pseudomonas* and *Alternaria* resistance in tobacco, besides smut and rust disease resistance in sugarcane.

## Lecture 11

### Application of biotechnology in plant disease management – Importance, production of pathogen free plants through tissue culture techniques

In modern terms “biotechnology” is defined as the manipulation, genetic modification and multiplication of living organisms through novel technologies, such as tissue culture and genetic engineering, resulting in the production of improved or new organisms and products that can be used in a variety of ways.

Genetic engineering is the technology by which it is possible to isolate particular genes from one organism, insert them into the genome of another organism and make them to express at right time. Cells of plants can be cultured in special nutrient medium and whole plants can be regenerated from cultured cells. This technique of growing plants *in vitro* is called “Tissue culture”.

In calli derived from infected tissues not all cells uniformly carry the pathogen. Only 40% of the single cells mechanically separated from TMV - infected tobacco callus contained the virus. The two possible reasons for the escape of some cells of a systemically infected callus from virus infection are-

- a) virus replication is unable to keep pace with cell proliferation, and
- b) some cells acquire resistance to virus infection through mutagenesis. Cells resistant to virus attack may even exist in the parent tissue together with susceptible ones. Several disease resistant plants have been evolved using somoclonal variation. Out of 370 tomato plants regenerated from calluses six showed resistant to TMV. Similarly, late blight (*Phytophthora infestans*) - resistant potato plants and bacterial blight of rice resistant calli have been evolved.

The pathogen produced secondary metabolites can be used to screen calluses for evolving disease resistant plants. Toxins will kill the calluses, but the mutant toxin resistant calluses will survive. The regenerated toxin resistant calluses yielded disease resistant plants. Brown spot pathogen (*Helminthosporium oryzae*) produced a host specific toxin for which resistant plants have been successfully developed. Similarly, *Helminthosporium maydis* - toxin resistant maize plants, *Phytophthora infestans* - resistant tobacco plants, *H. sacchari* resistant sugarcane plants have been evolved. Somaclonal variation refers to the tissue culture derived variation - Plants regenerated from somatic cells, using tissue culture, are not genetically uniform but displays significant genetic variability. This variability is very high when compared to spontaneous mutation. Somaclonal

variation has been demonstrated in a large number of plant species



(wheat, rice, oats, maize, tobacco, potato, sugarcane, brassica, etc.) for various traits such as resistance to fungal, viral and bacterial diseases. The procedure involves growing of cell cultures for several cycles on nutrient medium without any selective agent, followed by regeneration of plants.

The regenerants and their progenies are screened for disease resistance. Embryo rescue and protoplast fusion techniques are important to obtain hybrids among incompatible species and introgression of alien genes for disease resistance. In number of cases, useful genetic variability in the cultivated germplasm for resistance to diseases is either limited or lacking. Under such situations, wild germplasm seems to be a reservoir of useful genes for disease resistance. In the incorporation of alien genes, several crossability barriers are encountered. In many cases, the hybrid embryo aborts. However, the excised hybrid embryos when cultured on nutrient medium can be grown to plantlets. To incorporate alien genes from divergent sources, embryo rescue appears to be promising.

Tissue culture in conjunction with recombinant DNA technology is becoming increasingly important to insert foreign genes and produce transgenic plants. For successful infection of virus particles, the coat protein should be removed from viral RNA. If the host is made to synthesize coat proteins in large amount, naked viral RNA formation will be negligible. The host coat protein will encapsulate the RNA of the virus and prevent its multiplication. This will result in reduction and delay in symptom development. Eg. Transgenic tobacco plants expressing the tobacco mosaic virus coat protein protected the plants against this virus.

The expression of the viral genome in transgenic plants also conferred resistance to virus infection. These regions include portion of the viral replicase as well as, antisense RNA to coat protein. Transgenic tobacco plants transformed with a DNA copy of the satellite RNA of cucumber mosaic virus (CMV) were shown to produce large amounts of satellite RNA following inoculation with CMV and symptom development was greatly reduced.

Proteins with the ability to inhibit the growth of fungi *in vitro* are abundantly present in plants. Constitutive expression of these genes in transgenic plants may render these plants to fungus resistant. Transgenic tobacco plants constitutively expressing bean chitinase have been shown to display enhanced resistance to *Rhizoctonia solani*. Recently, tobacco plants expressing a ribosome-inactivating protein (RIP) from barley showed resistance to *R. solani*. The RIPs do

not inactivate self ribosomes and show activity towards ribosomes of distantly related species including those from fungi.

The constitutive expression of the groundnut stilbenesynthase gene in transgenic tobacco plants results in the synthesis of resveratrol (phytoalexin) and the transgenic plants show resistance to *Botrytis cinerea*.

Transgenic tobacco plants expressing acetyltransferase which detoxifies the tabtoxin, show resistance to *Pseudomonas syringae* pv. *tabaci*. More recently, chitinase gene from *Manduca sexta*, tobacco hornworm, has been cloned into *P. fluorescens* to increase their antagonistic potential against *R. solani*.

### **Meristem or shoot tip culture**

Meristem and shoot tip culture are used to eliminate virus from infected germplasm. It has long been observed that the rapidly growing meristems of plants are usually free of viruses, or at least have much lower concentration of viruses than nonmeristem cells. This situation has been exploited for the production of virus-free plants by meristem culture. It is commonly used in cassava, potato, sweet potato and ornamental plants.

“Virus-free” the term has been loosely used in literature. Plants infected with more than one type of virus and also may carry some unknown viruses. Thus, a plant can be claimed as free of only those viruses for which specific tests have given a negative result. However, the term “virus-free” is still retained by horticulturists for its commercial value.

Pathogen attack does not always lead to death of the plant. Many viruses may not even show visible symptoms. However, the presence of viruses in the plants can reduce the yield and quality of crops. It is well known that the distribution of viruses in plants is uneven. In infected plants, the apical meristems are generally either free or carry a very low concentration of the viruses. In the older tissues, the titre of the viruses increases with increasing distance from the meristem tips.

Five main possibilities have been suggested to explain the mechanisms underlying the „resistance“ of meristems to viruses.

- (i) Exclusion of the viruses from the meristems by lack of suitable vascular or plasmodesmal connections.
- (ii) Competition for key metabolites by the rapidly dividing meristem cells.
- (iii) The production of substances in meristem cells that result in breakdown of the virus.

- (iv) Deficiency in some key components of the machinery of virus replication, and
- (v) Presence of inhibitors of virus replication.

### **Factors affecting virus eradication**

Factors such as culture medium explant size and culture storage influence the virus eradication. In addition, heat treatment before or during culture significantly influences the efficiency of this technique. The physiological stage of the explants also affects virus elimination by meristem tip culture.

- (i) The success in obtaining complete plants can be considerably improved by the choice of the culture medium. The major features of the culture medium to be considered are its nutrients, growth regulators and physical nature.
- (ii) The size of meristem tip is an important factor governing regeneration capacity of meristems and to obtain virus free plants. For example, in cassava, meristems exceeding 0.2 mm size regenerated to plantlets, but those less than 0.2 mm size developed either Gallus or callus with roots. In general, the larger the meristem, the greater is the number of regenerated plants, but the number of virus free plants is inversely proportional to the size of meristem cultured.
- (iii) For meristem - tip cultures light incubation has generally proved better than dark incubation. The optimum light intensity for initiating tip cultures of potato is 100 lux, which should be increased to 200 lux after 4 weeks. The cultures are generally stored under standard culture room temperatures ( $25 \pm 2^\circ\text{C}$ ).
- (iv) Meristem tips should, preferably be taken from actively growing buds. Tips taken from terminal buds gave better results than those from axillary buds.

### **Meristem tip culture to eliminate Cassava Mosaic Virus**

Rapidly growing vegetative buds are excised, rinsed with sterile distilled water and then disinfected by immersing them in mercuric chloride solution (0.1%) for 2-3 minutes. The buds are then rinsed with several changes of sterile distilled water. Under the microscope, 3-4 leaf primordia (0.3 to 0.6 mm in size) is removed from the bud with a sterile scalpel. The buds are then aseptically transferred to Murashige and Skoog (MS) medium in test tubes and incubated at  $25 \pm 2^\circ\text{C}$  in light, for 45 days. The plantlets are then removed from the test tubes, washed in tap water and kept in Hoagland solution for 3-4 days for hardening. The plantlets are transferred to pots containing peat soil and vermiculite at 3:1 ratio and kept in mist chamber for 5-7 days. The plants are then transferred to glass house for further study.

### Disease Management by Biotechnological Methods

The use of genetically modified organisms and or modern techniques (genetic engineering, tissue culture etc.) with biological systems for disease control is known as biotechnology. Genetic engineering or Genetic manipulation is the deliberate alteration

of the composition of a genome by man. A growth of cells in a laboratory nutrient medium is known as tissue culture i.e. the technique of growing of plants in *vitro*. Cells of plants can be cultured in special nutrient medium and whole plants can be regenerated from cultured cells. Plant biotechnology is used for rapid clonal propagation of plants. It can help to produce industrial plant products under tissue culture conditions. Biotechnological methods are employed to control important plant diseases which are not amenable to control by usual methods.

#### Genetic Engineering

Genetic Engineering is the technology by which a particular gene is isolated from one organism and inserted into the genome of another organism and made to express at the right time.

#### Vectors for transfer of genes

Genetic engineering has been used to manage plant virus diseases. For transfer of genes to plants vectors are needed in which the gene to be transferred will multiply several folds. The most effective gene vector developed is the Tumor inducing plasmid of *Agrobacterium tumefaciens* from which the Tumor inducing genes have been removed. *A. tumefaciens* induces tumors (crown galls) through a Ti-plasmid (tumor-inducing) which is a circular double stranded DNA molecule containing up to 2,00,000 base pairs organized into several genes.

The Ti-plasmid is transferred from the bacterium into the cell. A specific region of the plasmid, the T-DNA, is transferred from the plasmid to the nucleus of the plant cell. It becomes integrated into the plant nuclear genome, and is transcribed. Cauliflower mosaic virus (CaMV) is the only plant virus with double-stranded DNA genome. As it has DNA genome, it is used as a possible vector in introducing foreign genes into plant. It is possible to insert a non-viral gene into CaMV genome and obtain expression of the gene in the infected plant. The viral promoter regions from CaMV are effective for

obtaining expression of other genes in plant cells. The genes to be expressed is now fused to a promoter element from CaMV and a gene of *A. tumefaciens*. They are then introduced into the plants using *A. tumefaciens* Ti-DNA transformation.

### **DNA construction**

Messenger RNA is extracted and exposed to an enzyme reverse transcriptase which synthesizes a complementary single stranded DNA. The complementary DNA (cDNA) is exposed to another enzyme, DNA polymerase, which produces the double stranded cDNA. The cDNAs are inserted into the plasmid of *A. tumefaciens*.

### **Coat-protein expression in transgenic plants**

Example: Transgenic tobacco plants expressing coat protein gene protected the plants against TMV. Transgenic tobacco plants showing resistance to alfalfa mosaic virus and tobacco rattle virus have also been developed. Transformation using a gene encoding the viral nucleocapsid protein of tomato spotted wilt virus (TSWV) has yielded transgenic tobacco plants that are resistant to TSWV. The expression of the viral genome in transgenic plants gives resistance to virus infection. Transgenic tobacco plants transformed with a DNA copy of the satellite RNA of cucumber mosaic virus (CMV) are shown to produce large amounts of satellite RNA following inoculation with CMV and symptom development is greatly reduced.

### **Satellite RNA expression in transgenic plants**

Satellite RNAs are associated with several viruses. They are packaged into virus particles along with the genomic RNAs of the helper virus. They are not part of the viral genome and have no obvious sequence relationships with the helper virus. The presence of the satellite RNA suppresses the disease severity in many hosts. Hence transgenic plants which express satellite RNA have been produced to manage virus diseases. e.g., Transgenic plants of tobacco expressed the synthesis of satellite tobacco ring spot virus and reduce the virus disease incidence. Satellite RNA expressing tobacco plants against Cucumber Mosaic Virus (CMV) and Tobacco etch virus have been synthesized.

### **MiRNA expression in transgenic plants**

A DNA copy is made of one or more sections of the viral genome that include the initiation codon for proteins vital to virus replication. The DNA copy is inserted in the host-cell genome, cells then produce an 'anti-sense RNA' called miRNA (mRNA-

interfering complementary to 5' end of the gene). The mic RNA hybridizes in vivo with the viral mRNA blocking translation. The mic RNA is inserted into the plants using the Ti plasmid of *A. tumefaciens*. Plants regenerated from the transformed cells will be resistant to the particular virus. This possibility is also being exploited for the control of virus diseases.

**Use of RFLP markers for cloning resistance genes**

Molecular markers viz., isozymes and DNA markers (Restriction Fragment Length Polymorphisms - RFLPs; Random Amplified Polymorphic DNA - RAPD and others) are being used in several areas relevant to identification of disease resistance genes. Some of the disease resistance genes using random DNA markers have been identified.

**Disease resistance genes mapped using RFLP markers**

<b>Plant</b>	<b>Pathogen</b>
Tomato	<i>Fusarium oxysporum</i>
Citrus	<i>Phytophthora</i> spp.

**Detoxification of pathotoxin**

Pathogens that produce pathogenesis-related phytotoxins usually also have the capacity to metabolize i.e. detoxify, these compounds. The search for genes encoding the enzyme(s) performing the key catabolic step(s) should thus lead to a convenient source of resistance, which can be engineered into plants to protect them from the effects of the toxin. A gene encoding a tabtoxin acetyltransferase from the pathogen, *Pseudomonas syringae* pv. *tabaci* which causes wild fire disease of tobacco was isolated and transferred into tobacco under a strong constitutive promoter. The transgenic plants expressed this gene and, when treated with either the pathogen or its toxin, did not produce the chlorotic lesion typical of wild fire disease.

**Activation of plant defense mechanism - Phytoalexins**

Phytoalexins have long been known to accumulate in certain plants upon infection by pathogens. The production of phytoalexins is also triggered by mechanical stimulation, ultraviolet (UV) irradiation, stress and a variety of chemical elicitors. Phytoalexins are part of the localized hypersensitive response at the site of damage or

pathogen ingress, which involves cell trauma and death. The importance of phytoalexins in the defense response is underscored by experiments and pathogenicity in *Nectria haematococca* was correlated to its ability to detoxify the phytoalexin, pisatin, by way of demethylation. By transferring the demethylase gene from *Nectria*, *Aspergillus nidulans*, a non-pathogen on peas, was rendered insensitive to pisatin.

### **Defense related genes**

#### **a. Single gene defense mechanism**

There are some defense proteins which do not require any intermediate step both for their synthesis and their expression require only few steps and those genes encoding such proteins are called single gene defense mechanism. Chitinases and glucanases are those proteins belonging to single gene defense mechanism.

#### **Chitinases and glucanases**

Chitinases are abundant proteins found in wide variety of plants. Although the physiological function of chitinases is not known, there is strong correlative evidence that they are defense proteins with antifungal activity. Chitin is a major structural component of cell walls of many fungi. The low constitutive activity of chitinase found in many plants can be dramatically induced by wounding or by infection of the tissue with fungal pathogens. Chitinase in concert with  $\beta$ -1,3-

glucanase (capable of degrading glucans present in fungal cell wall), degrades fungal cell walls and inhibits fungal growth at hyphal tips and has been shown to associate with hyphal walls in plants.

The chitinase and glucanase enzymes are having direct action against several fungal pathogens compared to other defense related proteins. Since lytic enzymes are encoded by single genes, these defense should be highly amenable to manipulation by gene transfer. The first reports of success with this approach was the expression of bean vacuolar chitinase gene under the control of the strong constitutive gene under the control of the strong constitutive promoter of the cauliflower mosaic virus (CaMV) 35S transcript into tobacco and *Brassica napus*, which resulted in decreased symptom development by *Rhizoctonia solani*, the causative agent of post-emergence damping off.

An endochitinase gene (from genomic tomato DNA library) was introduced into *Brassica napus* var. *oleifera*. The transgenic *Brassica* showed enhanced resistance against

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everal fungal pathogens like *Cylindrosporium concentricum*, *Phoma lingam* and



*Sclerotinia sclerotiorum* under field conditions when compared to non-transgenic plants. More recently, chitinase gene from *Manduca sexta*, tobacco horn worm, has been cloned into *P. fluorescens* to increase their antagonistic potential against *R. solani*.

### **b. Multigenic defense mechanism**

Defense responses such as phytoalexin biosynthesis or lignin deposition in the cell wall require the action of many genes.

#### **Peroxidases**

Anionic peroxidases in the cell wall catalyze the production of phenolic radicals for the oxidative polymerization of lignin from cinnamyl alcohols. In tomato, there is a marked induction of two linked genes encoding highly anionic peroxidases in an incompatible interaction with an avirulent form of *Verticillium albo-atrum*, with only weak induction in the compatible interaction with a virulent form of this vascular pathogen. Expression of one of these genes in transgenic tobacco under the control of either its own promoter or the CaMV35S promoter resulted in massive increase in anionic peroxidase activity and these plants apparently showed a significant increase in resistance to *Peronospora parasitica* as judged by symptom development and fungal sporulation.

#### **Activation of defense genes by chemicals**

Several classes of compounds have the potential to act as inducers of natural resistance mechanisms in horticultural crops and chemicals with such indirect modes of action may offer attractive alternatives or supplements to existing contact/systemic fungicides in integrated disease management. Increase was found to occur in response to salicylic acid treatment as well as oligosaccharides and glycoproteins originating from either fungal cell wall or host cell walls, the so called elicitors. Recently, chitosan seed treatment has been found to induce defense related genes like chitinase and glucanase in tomato and consequently the Fusarium crown and root rot diseases were significantly reduced. Pre-treatment with 2,6-dichloroisonicotinic acid was highly effective in significantly reducing both anthracnose (caused by *Colletotrichum lindemuthianum*) and rust (caused by *Uromyces appendiculatus*) diseases in bean plants.

**Cell and tissue culture**

Tissue culture approach is one of the oldest techniques in the field of molecular biology and it is applied in several ways for the development of disease resistance cultivars in agriculture and horticulture.

**a. Somaclonal Variation**

In the past two decades, several advances have been made in culturing of isolated plant cells and tissue under controlled conditions in vitro. When plants are regenerated from cultured cells, they exhibit new phenotypes, sometimes at high frequencies. If these are heritable and affecting desirable traits, such "somaclonal variation" can be incorporated into regular breeding programmes.

However, the finding of specific traits by these methods is largely left to chance and hence inefficient. Rather than relying on this undirected process, selection in vitro aims to specific traits by subjecting large populations of cultured cells to the action of a selective agent in the petri dish. For purpose of disease resistance, this selection can be done by fungal pathogens, culture filtrates of pathogens or isolated phytotoxins that are known to have a role in pathogenesis. The selection will allow only those cells to survive and proliferate that are resistant to the challenge. Plants regenerated from resistant cells often display a resistant phenotype when evaluated with either the toxin or the pathogen itself.

**Disease resistant plants from tissue culture**

Plant	Culture System	Selection	Resistance to Pathogen
Potato	Protoplasts	SCV	<i>Phytophthora infestans</i> <i>Alternaria solani</i>
	Callus	CF	<i>Fusarium oxysporum</i>
Tomato	Callus	Fusaric	<i>Fusarium oxysporum</i>
	Protoplasts	Acid	
Banana	Meristem	SCV	<i>Fusarium oxysporum</i>
Strawberry	Callus	SCV	<i>Fusarium oxysporum</i>

(SCV-plant regeneration without selection; CF crude culture filtrate)

Although this method has obviously yielded some impressive results, it also has its drawbacks; viz, i. Many pathogens do not produce pathogenesis specific toxins useful for selection ii. Culture filtrates are rather artificial and neither pathogens nor plant cells grown together in vitro behave quite as they would in a natural environment iii. This selection approach can only detect mutations in plant genes that are expressed at the time that selection is applied.

In order to be useful, new resistance traits, whether selected or not, must be heritable sexually or in the case of vegetatively propagated crops must be transmitted through vegetative propagules. The pathogens produced toxins can be used to screen calluses (cultured cells) which may regenerate resistant plants. The toxins will kill the calluses, but the mutant toxin resistant calluses will survive. The toxin-resistant calluses yield disease resistance plants. Vidhyasekaran obtained brown spot resistant rice plants using *Helminthosporium oryzae* toxin. Similarly, *H. maydis* resistant maize plants, *H. sacchari* resistant sugarcane plants and *Phytophthora infestans* resistant tobacco plants have been evolved.

#### **b. Anther culture**

In this method, the plants are produced directly from microspores (immature pollen grains). Through anther or microspore culture, one has immediate access to unique and rare combinations of genes representing the recombination of the genetic material contributed by the parents of the cross. Through anther culture, followed by chromosome doubling, such gene combinations can be fixed in their homozygous state as instant inbreds in a single step. Over the past two decades, anther culture has become widely accepted as a tool in cultivar development. This technique can be particularly useful for producing plants with novel combinations of resistance genes for managing fungal diseases.

#### **c. Protoplasmic fusion**

This generates hybrid cells by merging the total cellular components of somatic cells from which the cell walls have been removed to produce protoplasts. The incompatibility preventing sexual fertilization between species is thus avoided and viable hybrids have been created, even between unrelated distant species. Disease resistance genes have thus been transferred by protoplasts fusion from wild species into potato.

**INTEGRATED PLANT DISEASE MANAGEMENT (IDM) –  
CONCEPT, ADVANTAGES AND IMPORTANCE**

Integrated plant disease management can be defined as a decision-based process involving coordinated use of multiple tactics for optimizing the control of pathogen in an ecologically and economically. The implications are:

- ✓ Simultaneous management of multiple pathogens
- ✓ Regular monitoring of pathogen effects, and their natural enemies and antagonists as well
- ✓ Use of economic treatment thresholds when applying chemicals
- ✓ Integrated use of multiple, suppressive tactics.

**Principles of Plant Disease Control**

1. **Avoidance**—prevents disease by selecting a time of the year or a site where there is no inoculum or where the environment is not favorable for infection.
2. **Exclusion**—prevents the introduction of inoculum.
3. **Eradication**—eliminates, destroys, or inactivates the inoculum.
4. **Protection**—prevents infection by means of a toxicant or some other barrier to infection.
5. **Resistance**—utilizes cultivars that are resistant to or tolerant of infection.
6. **Therapy**—cure plants that are already infected

**Factors affecting occurrences**

Factors which affect plant diseases are micro-organisms, including fungi, bacteria, viruses, mycoplasmas, etc. or may be incited by physiological causes including high or low temperatures, lack or excess of soil moisture and aeration, deficiency or excess of plant nutrients, soil acidity or alkalinity, etc. Factors that limit the rate of disease development are the relatively low amounts of inoculum in the lag stage and the paucity of healthy plants available to the inoculum in the stationary stage.

The causative agents of disease in green plants number in the tens of thousands and include almost every form of life. But primary agents of disease may also be inanimate. Thus nonliving (abiotic) agents of disease include mineral deficiencies and excesses, air pollutants, biologically produced toxicants, improperly used pesticidal chemicals, and such other environmental factors as wind, water, temperature, and sunlight. Nonliving things certainly qualify as primary agents of disease; they continuously irritate plant cells and tissues; they are harmful to the physiological

processes of the plant; and they evoke pathological responses that manifest as the symptoms characteristic of the several diseases. But the abiotic agents of disease in plants. The abiotic agents of plant disease are termed noninfectious, and the diseases they cause are termed noninfectious diseases.

### **Micro-organisms**

The micro-organisms obtain their food either by breaking down dead plant and animal remains (saprophytes) or by attacking living plants and animals (parasites). In order to obtain nutrients, the parasitic organisms excrete enzymes or toxins and kill the cells of the tissues of the host plant, as a result of which either the whole plant or a part of it is damaged or killed, or considerable disturbance takes place in its normal metabolic processes.

### **Parasites**

One of the factors causing plant diseases is parasites, those living organisms that can colonize the tissues of their host-plant victims and can be transmitted from plant to plant. These biotic agents are, therefore, infectious, and the diseases they cause are termed infectious diseases. The infectious agents of plant diseases are treated in the standard textbooks on plant pathology.

### **Ability to produce an inoculum**

The parasitic pest must produce an inoculum, some structure that is adapted for transmission to a healthy plant and this can either parasitize the host directly or develop another structure that can establish a parasitic relationship with the host. For example, inocula for viruses are the viral particles (virions); for bacteria, the bacterial cells; for fungi, various kinds of spores or the hyphal threads of mold; for nematodes, eggs or second-stage larvae.

### **Agents/Media for transportation of inoculum**

The inoculum must be transported from its source to a part of a host plant that can be infected. This dispersal of inoculum to susceptible tissue is termed inoculation. Agents of inoculation may be insects (for most viruses and mycoplasma-like organisms and for some bacteria and fungi), wind (for many fungi), and splashing rain (for many fungi).

### **Wounds, Natural openings**

The parasite must enter the host plant, which it can do (depending on the organism) in one or more of three ways; through wounds, through natural openings, or by growing directly through the unbroken protecting surface of the host. Viruses are literally injected into the plant as the homopterous insect carrier probes and feeds within its host. Bacteria depend on wounds

or natural openings (for example, stomates, hydathodes, and lenticels) for entrance, but many fungi can penetrate plant parts by growing directly through plant surfaces, exerting enormous mechanical pressure and possibly softening host surfaces by enzymatic action.

### **Availability of food**

For occurrence of disease one of the factors affecting is, availability of nourishment to grow within its host. This act of colonization is termed infection. Certainly the parasite damages the cytoplasmic membranes of the host cells, making those membranes freely permeable to solutes that would nourish the parasite. And parasitism certainly results from enzymatic attacks by the parasite upon carbohydrates, proteins, and lipids inside the host cell. The breakdown products of such complex molecules would diffuse across the damaged host-cell membranes and be absorbed by the parasite in the form of sugars, amino acids, and the like. Air-borne parasites of foliage, flower, and fruit.

### **Preventive and control measures**

#### **A. PREVENTIVE**

#### **MEASURES Cultural practices**

Cultural practices usually influence the development of disease in plants by affecting the environment. Such practices are intended to make the atmospheric, edaphic, or biological surroundings favorable to the crop plant, unfavorable to its parasites. Cultural practices that lead to disease control have little effect on the climate of a region but can exert significant influence on the microclimate of the crop plants in a field. Three stages of parasite's life cycle namely, Survival between crops, production of inoculum for the primary cycle and inoculation can be controlled by following preventive measures.

#### **Survival between Crops**

Organisms that survive in the soil can often be controlled by crop rotations with unsuitable species. Depending on the system, either of two effects results. Catch crops have been used to control certain nematodes and other soil-borne pathogens. Soil-borne plant pathogens can be controlled by biological methods. Plant parasites may be controlled by antagonistic organisms that can be encouraged to grow luxuriantly by such cultural practices as green manuring and the use of appropriate soil additives. The soil-invading parasite thus becomes an amensal in association with its antagonist. Soil-borne plant parasites may also be killed during their over-seasoning stages by such cultural practices as deep ploughing (as for the

pathogen of southern leaf blight of corn), flooding (as for the cottony-rot pathogen and nematodes), and frequent cultivation and fallow (as for the control of weeds that harbor plantviruses). Plant diseases caused by organisms that survive as parasites within perennial hosts or within the seed of annual plants may be controlled therapeutically. Therapeutic treatments of heat and surgery are applicable here; those involving the use of chemicals will be mentioned later. Removal of cankered limbs (surgery) helps control fire blight of pears, and the hot-water treatment of cabbage seed controls the bacterial disease known as black rot. Heat therapy is also used to rid perennial hosts of plant-parasitic nematodes.

### **Production of Inoculum for the Primary Cycle**

Environmental factors (particularly temperature, water, and organic and inorganic nutrients) significantly affect inoculum production. Warm temperature usually breaks dormancy of overseasoning structures; rain may leach growth inhibitors from the soil and permit germination of resting spores; and special nutrients may stimulate the growth of overseasoning structures that produce inoculum.

### **Dispersal of inoculum and inoculation**

Cultural practices that exemplify avoidance are sometimes used to prevent effective dissemination. A second hierarchy of regulatory disease control is plant quarantine, the legally enforced stoppage of plant pathogens at points of entry into political subdivisions. The Plant Quarantine Act of the United States governs importation of plant materials into the country and requires the state govt. to enforce particular measures. Also, states make regulations concerning the movement of plant materials into them or within them. E.g., Florida imposes quarantine against the citrus-canker bacterium, which was eliminated from the state earlier by means of cooperative efforts led by the Florida Department of Agriculture.

### **Sample inspection**

One of the preventive measures to control the diseases is the use of sample inspection method. Laboratory evaluation of the representative sample drawn by the certification agency for the determination of germination, moisture content, weed seed content, admixture, purity, seed-borne pathogens.

## **B. Control**

### **Measures**

#### **Chemical Control**

The pesticidal chemicals that control plant diseases may be used in very different ways, depending on the parasite to be controlled and on the circumstances it requires for parasitic activities. E.g., a water-soluble eradicated spray is applied once to dormant peach trees to rid them of the overwintering spores of the fungus of peach-leaf curl, whereas relatively insoluble protective fungicides are applied repeatedly to the green leaves of potato plants to safeguard them from penetration by the fungus of late blight. Also, systemic fungicidal chemicals may be used therapeutically.

The oxathiin derivatives that kill the smut fungi that infect embryos are therapeutic, as is benomyl (which has systemic action against powdery mildews and other leaf infecting fungi). Volatile fungicides are often useful as soil-fumigating chemicals that have eradicated action. The chemical control of plant diseases is classified in three categories: seed treatments, soil treatments, and protective sprays and dusts.

#### **Seed Treatments**

Chemical treatments of seed may be effective in controlling plant pathogens in, on, and around planted seed. Seed treatment is therapeutic when it kills bacteria or fungi that infect embryos, cotyledons, or endosperms under the seed coat, eradicated when it kills spores of fungi that contaminate seed surfaces, and protective when it prevents penetration of soil-borne fungi into seedling stems. Certified seed is usually given treatment necessary for the control of certain diseases. Seed treatment is of two types; viz., physical and chemical. Physical treatments include hot-water treatment, solar-heat treatment (loose smut of wheat), and the like. Chemical treatments include use of fungicides and bactericides. These fungicides are applied to seed by different methods. In one method, the seed in small lots is treated in simple seed-treaters. The seed-dip method involves preparing fungicide suspension in water, often at field rates, and then dipping the seed in it for a specified time.



**Some chemicals commonly used to control plant diseases**

Chemical and use	Relative toxicity	
	Oral	Dermal
<b>Seed treatments (all fungicides)</b>		
Chloraneb	Low	Low
Dichlone	Low	High
Thiram	Moderate	High
Carboxin (systemic and therapeutic)	Low	Low
<b>Soil treatments</b>		
Methyl bromide <sup>b</sup> (general pesticide)	Very high	Very high
PCNB (fungicide)	Low	Moderate
SMDC [vapam] (fungicide, nematocide)	Moderate	Moderate
MIT ["Vorlex"] (fungicide, nematocide)	Moderate	Moderate
D-D mixture (nematocide)	Moderate	Low
<b>Plant-protective treatments</b>		
Copper compounds (fungicides, bactericides)	Moderate	Low
Sulfur (fungicide)	Low	Moderate
Maneb (fungicide)	Very low	Low
Zineb (fungicide)	Very low	Low
Captan (fungicide)	Very low	Very low
Dinocap (fungicide for powdery mildews)	Low	Low
Streptomycin (bactericidal antibiotic)	Very low	Low
Cyclohexamide <sup>b</sup> (fungicidal antibiotic)	Very high	Very high
Benomyl (protective and therapeutic fungicide)	Very low	Very low

The oxathiins (carboxin, DMOC) used to kill embryo infecting smuts of cereal grains have little effect on other organisms, most eradicative and protective chemicals have a wider range of fungicidal activity; they are effective against most seed-infesting and seedling-blight fungi. But specific seed-treatment chemicals often work best to control lagivendiseases of a

single crop-plant species. Moreover, the toxicity of chemicals to seeds varies, and farmers should use only the compounds recommended by the Cooperative Extension Service of their country and state.

Copper and mercury-containing compounds were first used as seed-treating chemicals. But copper is toxic to most seeds and seedlings, and mercury has been banned from use in seed treatments because of the danger it poses to humans and animals. Organic compounds now widely used as protective and eradicated seed treatments include thiram, chloraneb, dichlone, dexton, and captan.

### **Soil Treatments**

Soil-borne plant pathogens greatly increase their populations as soils are cropped continuously, and finally reach such levels that contaminated soils are unfit for crop production. Chemical treatments of soil that eradicate the plant pathogens therein offer the opportunity of rapid reclamation of infested soils for agricultural uses. Preplanting chemical treatment of field soils for the control of nematode-induced diseases, and fumigation of seedbed and greenhouse soils (with methyl bromide, for example) is commonly practiced to eradicate weeds, insects, and plant pathogens. Field applications of soil-treatment chemicals for fungus control are usually restricted to treatments of furrows. Formaldehyde or captan applied is effective against sclerotia-producing fungi that cause seedling blights, stem rots, and root rots of many field crops. Other soil-treatment fungicides are vapam and "Vorlex." Soil treatments made at the time of planting are most effective against parasitic attacks that come early in the growing season.

### **Protective sprays and dust**

Protective fungicides prevent germination, growth, and penetration. In order to use protective fungicides effectively, the farmer must not only select the right fungicide for the job, but also apply it in the right amount, at the right times, and in the right way. Too little fungicide fails to control disease; too much may be toxic to the plants to be protected. The farmer and applicator, therefore, must always follow use instructions to the letter. Timing of applications is also critical.

### **Advantages**

Integrated approach integrates preventive and corrective measures to keep pathogen from causing significant problems, with minimum risk or hazard to human and desirable components of their environment.

Some of the benefits of an integrated approach are as follows:

- Promotes sound structures and healthy plants
- Promotes the sustainable biobased disease management alternatives.
- Reduces the environmental risk associated with management by encouraging the adoption of more ecologically benign control tactics
- Reduces the potential for air and groundwater contamination
- Protects the non-target species through reduced impact of plant disease management activities.
- Reduces the need for pesticides and fungicides by using several management methods
- Reduces or eliminates issues related to pesticide residue
- Reduces or eliminates re-entry interval restrictions
- Decreases workers, tenants and public exposure to chemicals
- Alleviates concern of the public about pest & pesticide related practices.
- Maintains or increases the cost-effectiveness of disease management programs

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