

**DISEASES OF FIELD AND HORTICULTURAL CROPS AND THEIR  
MANAGEMENT - II**

**PRACTICAL MANUAL**

**Course Code: CC-AGP646**



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## Preface

This course will be helpful to clear concept of the students on detection and diagnosis of important diseases of field and horticultural crops by their symptoms. This Practical manual will also provide practical knowledge on methods for microscopic slide preparation (Teasing method) and Lactophenol Cotton Blue (LPCB) staining, free hand section method for histopathology study, methods for preparation of Herbarium specimens of plant disease sample, identification and histopathological studies of different field and horticulture plant diseases viz. Black rust of Wheat, Brown rust of Wheat, White rust of Mustard, Alternaria blight of Mustard, Downy mildew of Cucurbits, Stemphylium blight of Onion, Anthraenose of Chilli, Leaf spot of Turmeric, Stem gall of Coriander, Botrytis blight of Marigold, Black spot of Rose, Late blight of Potato, and Early blight of Potato.

This course will create awareness on diseases of Field and horticultural crops not only reduce the productive capability but also adversely affect the quality. With the changing climate, Agricultural & horticultural crops have become vulnerable to hazards of diseases, playing a major role in causing threat of food security.

By the end of this course, the students will be able to know the distribution of plant diseases and their economic importance identify the diseases based on the symptomatology and the factors influencing the disease development, and acquaint themselves with integrated disease management practices.

### Practical (P) 1: Field visits for Detection and diagnosis of Field Crop Diseases

| Diseases of Field Crops |                     |                 |          |
|-------------------------|---------------------|-----------------|----------|
| Sl. No.                 | Name of the Disease | Causal Organism | Symptoms |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
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| Diseases of Field Crops |                     |                 |          |
|-------------------------|---------------------|-----------------|----------|
| Sl. No.                 | Name of the Disease | Causal Organism | Symptoms |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |
|                         |                     |                 |          |

**Practical (P) 2: Field visits for Detection and diagnosis of Horticultural Crop Diseases**

| Diseases of Horticultural Crops |                     |                 |          |
|---------------------------------|---------------------|-----------------|----------|
| Sl. No.                         | Name of the Disease | Causal Organism | Symptoms |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |

| Diseases of Horticultural Crops |                     |                 |          |
|---------------------------------|---------------------|-----------------|----------|
| Sl. No.                         | Name of the Disease | Causal Organism | Symptoms |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |
|                                 |                     |                 |          |

### Practical (P) 3: Methods for preparation of Herbarium specimens of plant disease sample

**Objectives:** Dry preservation of plant disease samples.

A Herbarium is defined as a collection of plants that usually have been dried, pressed, preserved on sheets and arranged according to any accepted system of classification for future reference and study.

In fact, it is a great filing system in Plant Pathology for information about plant disease, symptoms, both primary in the form of actual specimens of the plants, and secondary in the form of published information, pictures and recorded notes.

**Materials required:** Knife, Scissor, Plant disease sample, Old newspaper or blotting paper, Adhesive glue or cello-tape, Label, Weight for proper pressing

**Methods for preparation of a herbarium:**

1. **Collection of plant disease sample:** Disease sample should be collected from fields or gardens and packed in polypropylene bag.
2. **Pressing and drying:** The specimens are spread out between the folds of old newspapers or blotting sheets avoiding overlapping of parts. The blotting sheets with plant disease specimen should be placed in the plant press for drying. After 24 to 48 hrs the press is opened. The process should be repeated for proper drying.
3. **Mounting:** The dried specimens are mounted on herbarium sheets of standard size (41 x 29 cm). Mounting is done with the help of glue, adhesive or cello-tape.
4. **Labelling:** A label is pasted or printed on the lower right hand corner. The label should indicate the information about the name of disease, host (common and scientific name), causal organism, date of collection, place of collection and name of collector.



Potato late blight herbarium specimen



Citrus canker herbarium specimen



Rust disease herbarium specimen

Useful internet links: <https://youtu.be/xj7KQR6ck5g> , <https://youtu.be/6MDfQjAOxes>



## Practical (P) 4: Identification and histopathological studies of Black rust of Wheat

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** The stem rust disease caused by fungus *Puccinia graminis* f. sp. *tritici* appears as elongate blister like pustules, most frequently on the leaf sheaths of the wheat plant, but also on stem tissues, leaves, glumes, and awns. Stem rust pustules on leaves develop mostly on the lower side, but may penetrate and make limited sporulation on the upper side. As infected plants mature, uredinia change into telia, altering colour from red into dark brown to black, thus the disease is also called black rust. Severe infection of stems interrupts nutrient flow to the developing heads, resulting in shriveled grains and stems weakened by rust infection are prone to lodging.



Symptoms of Black/ Stem rust of Wheat

**Causal organism:** *Puccinia graminis* f. sp. *tritici* (**Division:** Basidiomycota, **Class:** Pucciniomycetes, **Order:** Pucciniales, **Family:** Pucciniaceae)

It is a macrocyclic, heteroecious rust pathogen that produces five different spores (Urediniospores, Teliospores and Basidiospores on Wheat and Pycniospores and Aeciospores on Barberry).

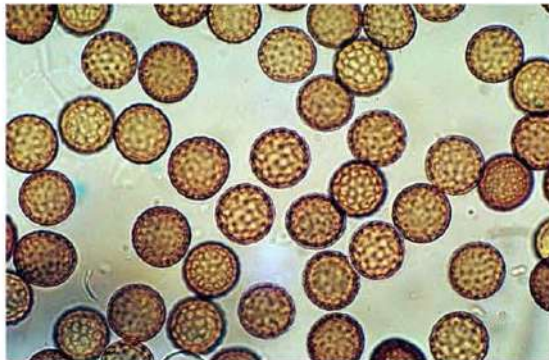
Plants do not usually show obvious disease symptoms until 7 to 15 days after infection when the oval pustules (uredinia) of powdery, brick-red **urediniospores** break through the epidermis. Microscopically, these red spores are covered with fine spines.

Microscopically, **teliospores** are two celled and thick walled.

**Basidiospores** are haploid, unicellular, thin walled, short lived sexual spores produced exogenously on basidium or epibasidium (promycelium).

Pycnia appear on barberry plants in the spring, usually in the upper leaf surfaces. They are often in small clusters and exude **pycniospores** in a sticky honeydew. Five to 10 days later, cup-shaped structures filled with orange-yellow, powdery **aeciospores** break through the lower leaf

surface. The aecial cups are yellow and sometimes elongate to extend up to 5 mm from the leaf surface. Microscopically, aeciospores have a slightly warty surface.



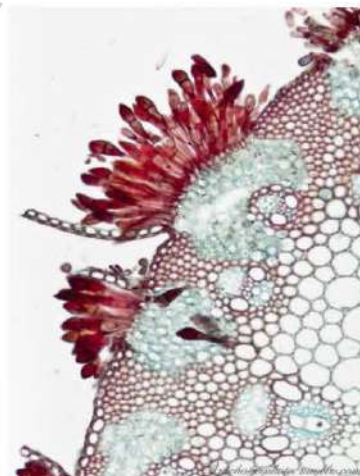
**Urediniospores**



**Teliospores**



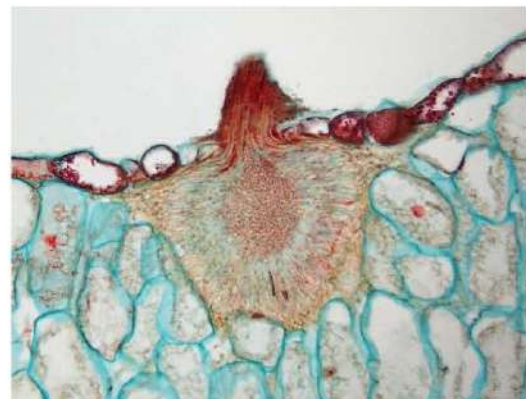
**Aecium with aeciospores**



**Telium with Teliospores**



**Promycelium with basidiospores**



**Pycnium with pycniospores**



**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Razor blade, Fine pointed needle, Brush, Watch glass, Forceps, Carrot disc, Cotton blue, Lactophenol, Sterile water, Microscope

**Methods:**

1. Obtain a new double edge razor blade. To minimize the risk of cutting oneself, cover one edge of the razor blade with masking tape.
2. Insert the thin diseased leaves, into a small piece of pith such as a carrot disc.
3. Hold piece of carrot firmly. The material should be held against the side of the first finger of the left hand (or right hand) by means of the thumb.
4. Take the razor blade in the right hand (or left hand) and place it on the first finger of the left hand (or right hand), more or less at a right angle to the specimen. Draw the razor across the top of the material in such a way as to give the material a drawing cut (about 45° in the horizontal direction).
5. Cut several sections at a time. Sections will certainly vary in thickness.
6. Transfer sections to water, always using a brush, not a forceps or needle.
7. Select and transfer the thinnest section and stain with LPCB.

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/AeuP5IYP5HA>

## Practical (P) 5: Identification and histopathological studies of Brown rust of Wheat

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** Symptoms include pustules which are circular or slightly elliptical, smaller than those of stem rust, usually do not coalesce, and contain masses of orange to orange-brown urediospores. Infection sites primarily are found on the upper surfaces of leaves and leaf sheaths and occasionally on the neck and awns.



Symptoms of Brown rust of Wheat

**Causal organism:** *Puccinia triticina* (**Division:** Basidiomycota, **Class:** Pucciniomycetes, **Order:** Pucciniales, **Family:** Pucciniaceae)

It is a macrocyclic, heteroecious rust pathogen that produces five different spores (Urediniospores, Teliospores and Basidiospores on Wheat and Pyreniospores and Aeciospores on *Thalictrum*).

Urediniospores are round shaped with 7-10 scattered germ pores, 20-28µm sized.

Teliospores are dark brown, two-celled with thick walls and rounded or flattened at the apex.



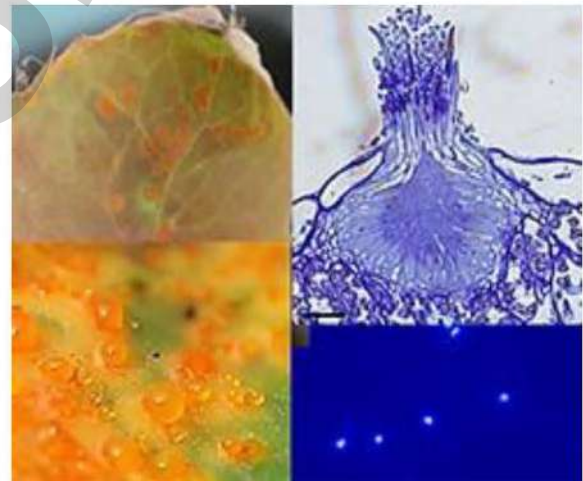
**Urediniospores**



**Teliospores**



**Aecium with aeciospores**



**Pycnium with pycniospores**

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Razor blade, Fine pointed needle, Brush, Watch glass, Forceps, Carrot disc, Cotton blue, Lactophenol, Sterile water, Microscope

**Methods:** Free hand sectioning method followed by lactophenol cotton blue staining [See Practical (P) 4]



**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/piqLPv9PCyE>

## Practical (P) 6: Identification and histopathological studies of White rust of Mustard

**Objectives:** To identify the causal organism of the disease and to study etiology.

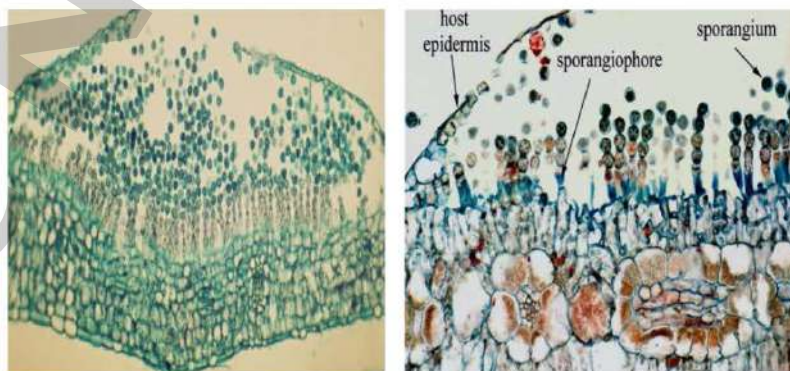
**Disease symptoms:** White or creamy yellow pustules of various shape and size appear on the surface of the leaves, mainly on the lower surface. In severe infection the leaves become thick, fleshy, inrolled and their size becomes reduced. If young stems and inflorescence are infected the fungus becomes systemic inside tissues and produces deformities like swelling and distortion of the floral parts mainly due to hypertrophy and hyperplasia forming a 'staghead structure'.



Symptoms of White rust of mustard

**Causal organism:** *Albugo candida* (Phylum: Oomycota, Order: Albuginales, Family: Albuginaceae)

Mycelium of fungus is intercellular forming knob like haustoria in the host cells. Hyphae forms beneath the epidermis and it gives rise to the sporangial beds or sori. Sporangiphores arise from the sori are free from each other laterally and are very thick towards the base. Sporangia are formed in basipetal succession in chains are hyaline, and spherical and produces zoospores.



Sporangiphore and sporangia at 10X

Sporangiphore and sporangia at 40X

*Albugo candida*: sporangia and sporangiphore

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Razor blade, Fine pointed needle, Watch glass, Carrot disc, Cotton blue, Lactophenol, Sterile water, Microscope

**Methods:** Free hand sectioning method followed by lactophenol cotton blue staining [See Practical (P) 4]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/j0yfbFa3Zik>



## Practical (P) 7: Identification and histopathological studies of Alternaria blight of Mustard

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** The fungus attacks the lower leaves as small circular brown necrotic spots and slowly increases in size. In severe cases many concentric spots coalesce to cover large patches showing blighting and defoliation. Circular to linear dark brown spots also develop on stems and pods which become elongated at later stage. Infected pods produce small discolored and shriveled seeds.



Symptoms of Alternaria blight mustard

**Causal organism:** *Alternaria brassicae* (**Division:** Ascomycota, **Class:** Dothideomycetes, **Order:** Pleosporales, **Family:** Pleosporaceae)

Conidia are the asexual spores formed. They are formed in chains or solitary, typically ovoid to obclavate, often beaked, pale brown to brown, multi-celled and muriform.



Conidia of *Alternaria brassicae*

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:**

- i. Place a drop of cotton blue on the slide.
- ii. Cut a small portion of the infected samples (lesion) and put on the mounting fluid.
- iii. Wait for one minute for getting stain the sample and after that wash the cotton blue with the help of Lacto phenol.
- iv. Scrap the disease portion from the infected leaf.
- v. Place the cover slip slowly at an angle with support of the needle on the drop of the mounting fluid.
- vi. Dry excess of lacto phenol with a small piece of blotting paper.
- vii. Expel air bubbles, if any under the cover slip.
- viii. Examine the sample at low power (10X) then at high power (40X) of the microscope.

**Observation:****Result and conclusion:**

**Useful internet links:** <https://youtu.be/rmIyBI2V5xA>

## Practical (P) 8: Identification and histopathological studies of Downy mildew of Cucurbits

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** The disease is most severe on cucumber and muskmelon but it can infect all cucurbit crops. Symptoms occur mainly on the leaves as yellow angular spots restricted by veins and becoming tan to brown with age. In high moisture, sporulation may be evident to the naked eye on the lower surface of leaf lesions. Under extremely heavy infection, leaves become necrotic followed by death of the whole plant.



Symptoms of Downy mildew disease of cucurbits

**Causal organism:** *Pseudoperonospora cubensis* (**Phylum:** Oomycota, **Order:** Peronosporales, **Family:** Peronosporaceae)

Sporangiophores with dichotomously branched structure with pointed tips and sporangia germinate through the release of zoospores.



*Pseudoperonospora cubensis*: sporangia with sporangiophore



**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process followed by lactophenol cotton blue staining [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/sz0vZ-t0gvg>

## Practical (P) 9: Identification and histopathological studies of *Stemphylium* blight of Onion

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** The main symptoms of the disease appear in the middle of the leaf as small, yellow to pale orange flecks or streaks which later develop into elongated spindle shaped diffused spots surrounded by characteristic pinkish margin. The spots turn grey at the center and later become brown to dark brown with the appearance of conidiophores and conidia of the fungus. Gradually the entire foliage is blighted. Similar symptoms also develop on inflorescence stalks.



Symptoms of *Stemphylium* blight of Onion

**Causal organism:** *Stemphylium vesicarium* (*Pleospora allii*) (**Phylum:** Ascomycota, **Class:** Dothidiomycetes, **Order:** Pleosporales)

Conidiophores are macronematous, mononematous, scattered, un-branched, straight or flexuous, cylindrical, smooth, septate, pale to mid brown,  $(30-47-52 (-67) \times 4-5 \mu\text{m})$ . Conidiogenous cells are monoblastic, integrated, terminal, swollen, dark brown,  $6-7 \mu\text{m}$  diam, with up to 3 proliferations. Immature conidia are ellipsoid, rounded at the ends, mature conidia are  $20-24 (-30) \times 12-15 \mu\text{m}$ , with length/width ratios approaching 1.5–2.3, solitary, acrogenous, oblong to broadly ellipsoid, sub-truncate basally, rounded to sub-truncate apically, golden-brown to olive-brown, with 1–3 transverse and 1–4 longitudinal or oblique septa, often constricted at one or more of the septa, cicatrized at the base, with a verrucose conidial wall.



Conidia of *Stemphylium vesicarium* under 10X and 40X

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/0sxuY7qyRU>



## Practical (P) 10: Identification and histopathological studies of Anthracnose of Chilli

**Objectives:** To identify the causal organism of the disease and to study etiology.

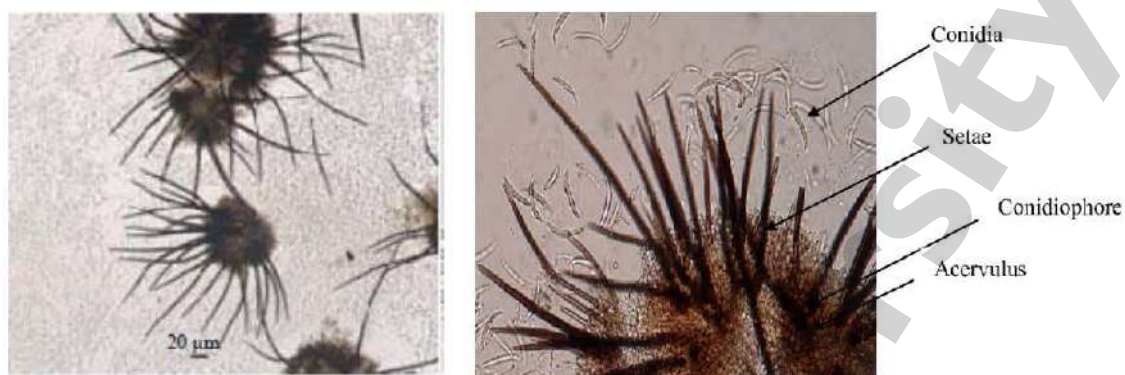
**Disease symptoms:** Ripe fruits turning red are affected. Small, black, circular spot appears on the fruit skin. Badly diseased fruits turn straw colour or pale white colour, lose their pungency. Necrotic tissues appear greyish white with black dot like acervuli. In advanced stage, seeds covered by a mat of fungal hyphae, turn rusty in colour.



Symptoms of Chilli anthracnose

**Causal organism:** *Colletotrichum capsici* (**Phylum:** Ascomycota, **Class:** Sordariomycetes, **Order:** Glomerellales, **Family:** Glomerellaceae)

The mycelium is septate and grows both inter and intra cellularly in the host tissue. The asexual fruiting bodies, acervuli contain many rigid, brown coloured, 1-5 septate setae. A large number of conidia are borne on conidiophores in each acervulus which are falcate, unicellular, hyaline having a normally truncated base.



*Colletotrichum capsici*: Acervuli and conidia under 10X and 40X

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/7riyluoQd9A>

## Practical (P) 11: Identification and histopathological studies of Leaf spot of Turmeric

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** On leaves, elliptic to oblong spots appear on both the surfaces, but more on upper surface. Mature spots have grayish center with dark brown margins surrounded by a yellow halo. Central portion of the spot becomes thin and papery. Several spots coalesce to form irregular necrotic patches.



Symptoms of Turmeric leaf spot

**Causal organism:** *Colletotrichum capsici* (Phylum: Ascomycota, Class: Sordariomycetes, Order: Glomerellales, Family: Glomerellaceae)

The mycelium is septate and grows both inter and intra cellularly in the host tissue. The asexual fruiting bodies, acervuli contain many rigid, brown coloured, 1-5 septate setae. A large number of conidia are borne on conidiophores in each acervulus which are falcate, unicellular, hyaline having a normally truncated base.



*Colletotrichum capsici*: Acervuli and conidia



**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/TeseFvFRJSA>

## Practical (P) 12: Identification and histopathological studies of Stem gall of Coriander

**Objectives:** To identify the causal organism of the disease and to study etiology.

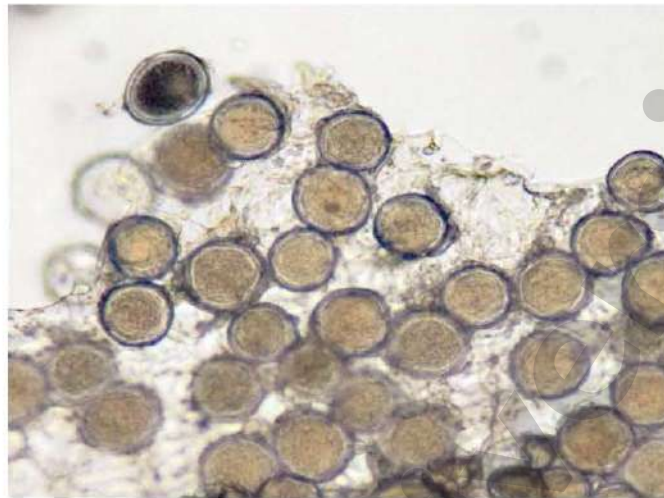
**Disease symptoms:** The disease manifests itself in the form of small tumour-like swellings on all herbaceous parts of the affected plants, namely, stems, petioles, leaf stalk, peduncles, flower stalks, fruits and leaves. The swellings are usually elongated and 912 mm x 3-5 mm in diameter, the size varies according to the dimension of the part infected. The fungus is restricted to tumours only. The hyperplasy of the outer cortical cells restricted to those in the neighbourhood of intercellular hyphae results in the formation of turnours. Very often the thin-walled parenchymatous cortical cells may replace the collenchyma in the hypertrophied tissue. The infection becomes systemic in host stem before or during pre-flowering period, inducing organogenic changes in flowers and fruits due to hypertrophy and hyperplasy.



Symptoms of Stem gall of Coriander

**Causal organism:** *Protomyces macrosporus* (**Phylum:** Ascomycota, **Class:** Taphrinomycetes, **Order:** Thaprinales, **Family:** Protomycetaceae)

The Hyphae are intercellular, closely septate and broad; branching is irregular, scattered cells in the hyphae swell, form ellipsoidal or globose bodies, which later develop in to chlamydospores. As the chlamydospores mature, a thick, hyaline and three- layered wall measuring 50 to 60µm in diameter surrounds them. The mycelium of the fungus is only found in the tumours although the resting spores of the fungus cause systemic infection.



Chlamydospores of *Protomyces macrosporus*

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Razor blade, Fine pointed needle, Brush, Forceps, Watch glass, Carrot disc, Cotton blue, Lactophenol, Sterile water, Microscope

**Methods:** Free hand sectioning method followed by lactophenol cotton blue staining [See Practical (P) 4]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/H49Jo-OeMGw>



## Practical (P) 13: Identification and histopathological studies of Botrytis blight of Marigold

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** Flower parts develop water-soaked lesions that become necrotic and die. A gray mass of spores develops on necrotic tissue during wet conditions. Entire petals or flowers may become diseased. Senescent leaves may also be attacked. Reduce the aesthetic value of the flowers.



Symptoms of Botrytis blight of Marigold

**Causal organism:** *Botrytis cinerea* (**Phylum:** Ascomycota, **Order:** Helotiales, **Family:** Sclerotiniaceae), fungus that colonizes dead, dying, and wounded plant parts.

Conidia from naturally infected flowers are ellipsoidal or ovoid,  $6.1$  to  $8.5 \times 5.1$  to  $9.8 \mu\text{m}$ . Conidiophores from PDA (potato dextrose agar) cultures are straight or flexuous, septate, with an inflated basal cell brown to light brown, and measured  $105$  to  $425 \times 9$  to  $28 \mu\text{m}$ . After 21 days, the fungus forms black sclerotia ranging from  $0.7$  to  $4.8 \times 1.0$  to  $3.7 \text{ mm}$ .



Conidia and conidiophores of *Botrytis cinerea*

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/VLgdtA9I0ec>

## Practical (P) 14: Identification and histopathological studies of Black spot of Rose

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** Circular black spots, frequently with fringed (diffuse or feathery) margins, on the leaf. Yellowing and defoliation are common in susceptible cultivars. Bare stems with few leaves attached near the top is a frequent symptom. In wet weather, spots may become very severe and coalesce to make large irregular spots covering a third of the leaf surface. In cases of severe infections, similar appearing lesions also form on the stems.



Symptoms of black spot of rose

**Causal organism:** *Diplocarpon rosae* (Asexual stage: *Marssonina rosae*) (**Phylum:** Ascomycota, **Order:** Helotiales, **Family:** Dermateaceae)

The vegetative body of the fungus consists of two parts viz., the subcuticular mycelium and the internal mycelium. The fungus produces acervuli on the central part of the tar spots as blister like projections. Conidiophores are hyaline short and cylindrical. Conidia are hyaline, two celled, fusiform or allantoid to obelavate, upper end round, base narrow, guttulate,  $18 - 25 \times 5 - 6 \mu\text{m}$ .



*Marssonina rosae*: acervular conidiomata and conidia



**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/G1K2LYuOnGA>

## Practical (P) 15: Identification and histopathological studies of Late blight of Potato

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** Circular Brown to purplish black water soaked lesions covers the entire leaf, petiole and stem. Enlarge rapidly, lower surface shows whitish mildew growth, severe defoliation may occur. Potato tuber: purplish slightly sunken lesions leading to dry rot. Rotten portion emits characteristic odour.



Symptoms of Potato late blight

**Causal organism:** *Phytophthora infestans* (**Phylum:** Oomycota, **Order:** Peronosporales, **Family:** Peronosporaceae)

Mycelium is endophytic, coenocytic, hyaline, branched, inter-cellular. Haustoria club shaped. Sporangiophores are hyaline with nodular swellings, branched, indeterminate, thick walled, arise through stomata on leaves or lenticels on tubers. Sporangia are multinucleate, thin-walled, hyaline, oval or pear or lemon shaped with a definite papilla at the apex.



*Phytophthora infestans*: Sporangia with sporangiophore, zoospores

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/jANKhuBXvqk>, <https://youtu.be/tySLhhYb70k>



## Practical (P) 16: Identification and histopathological studies of Early blight of Potato

**Objectives:** To identify the causal organism of the disease and to study etiology.

**Disease symptoms:** The disease appears on the leaflets, 3-4 weeks after the crop is sown as small, isolated, scattered pale brown to dark spots, oval or angular in shape mostly up to 3 or 4 mm in diameter. Each spot is usually delimited by a narrow chlorotic marginal zone which fades into the normal green. The chlorotic zone increases with the increase in size of the spot. The necrotic tissue of the spot often shows a series of concentric ridges which produce a target-board effect, a symptom characteristic of this disease. The number of spots on the leaflets may be a few but if the conditions are favourable the spots increase in number and size involving the entire leaf surface. Falling of leaves starts with the older (lower) ones until a few remain at the top.



Symptoms of Potato early blight

**Causal organism:** *Alternaria solani* (**Division:** Ascomycota, **Class:** Dothideomycetes, **Order:** Pleosporales, **Family:** Pleosporaceae)

The mycelium consists of light brown, slender, septate sparsely branched hyphae which become dark-coloured with age. The hyphae ramify in the intercellular spaces but later penetrate the cells of the invaded tissues. The conidiophores which are relatively short (50- 90  $\mu$  long and 9  $\mu$  broad) and dark-coloured arise from the older diseased tissue of the host and emerge through the stomata. The conidia which measure 120 to 296  $\mu$  in length and 12-20  $\mu$  in breadth are dark-coloured, beaked, muriform and multiseptate.



Conidia of *Alternaria solani*

**Materials required:** Infected disease sample, Slide, Cover slip, Blotting paper, Fine pointed needle, Scissor, Cotton blue, Lactophenol, Microscope

**Methods:** Teasing process [See Practical (P) 7]

**Observation:**

**Result and conclusion:**

**Useful internet links:** <https://youtu.be/VK-r3rdvzaE>, <https://youtu.be/eytqKPFZ6sw>