



# Purnima International School

Shree Swaminarayan Gurukul, Zundal

Class: XII Science

Subject: Biology Practical

Experiments (2022-23)

| Exp. No          | Aim  |
|------------------|--|
| <b>Section-A</b> |  |
| 1                | Prepare a temporary mount to observe pollen germination  |
| 2                | Study the plant population density by quadrat method   |
| 3                | Study the plant population frequency by quadrat method.  |
| 4                | Prepare a temporary mount of onion root tip to study mitosis.  |
| 5                | Isolate DNA from available plant material such as spinach, green pea seeds, papaya, etc.   |
| <b>Section-B</b> |  |
| 6                | Flowers adapted to pollination by different agencies (wind, insects, birds)  |
| 7                | Identification of stages of gamete development, i.e., T.S. of testis and T.S. of ovary through permanent slides (from grasshopper/mice)  |
| 8                | Meiosis in onion bud cell or grasshopper testis through permanent slides   |
| 9                | T.S. of blastula through permanent slides (Mammalian)  |
| 10               | Mendelian inheritance using seeds of different colour/sizes of any plant   |
| 11               | Prepared pedigree charts of any one of the genetic traits such as rolling of tongue, blood groups, ear lobes, widow's peak and colour blindness  |
| 12               | Controlled pollination - emasculation, tagging and bagging   |
| 13               | Common disease causing organisms like Ascaris, Entamoeba, Plasmodium, any fungus causing ringworm through permanent slides, models or virtual images or specimens. Comment on symptoms of diseases that they cause |

## Experiment-1

**Aim:** To Observe pollen germination by preparing temporary mount.

### **Material/Apparatus Required:**

- Fresh mature flower like *Tradescantia* jasmine, lily, Vinca, China Rose, *Petunia*, etc.
- Glass slide
- Glass rod
- Sucrose
- Potassium nitrate
- Boric acid
- Compound microscope
- Cover slip
- Beaker
- Distilled water
- Dropper
- Magnesium sulphate

### **Principle:**

**Pollen Grains:** Pollen grains represent the male gametophytes. For example, if the open anthers of any flower is seen, yellowish powdery pollen are found. They are generally spherical in shape about 25-50 micrometers in diameter. It has a prominent two-layered wall. The hard outer layer called the exine is made up of sporopollenin. The exine has prominent apertures called germ pores. The inner wall of pollen grain is called the intine. It is a thin and continuous layer made up of cellulose and pectin.

**Pollen Grains Germination:** In nature, pollen grains germinate on the compatible stigmas of carpels. This process is called pollen pistil interaction. They can also induce to germinate in a synthetic medium using chemical which favour pollen germination such as 10% sucrose solution.

During germination, intine (inner wall) of pollen grain emerges out as pollen tube through one of the germ pores in exine (Outer wall).

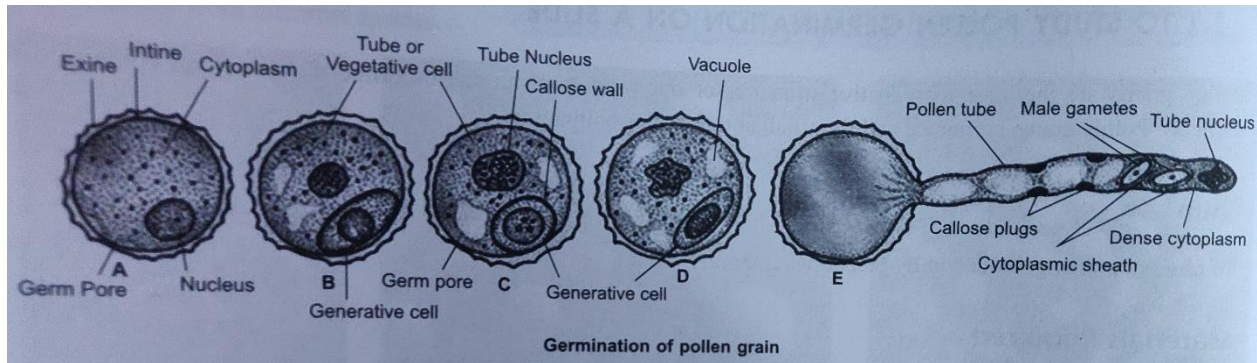
### **Procedure:**

- Prepare the medium for pollen germination by dissolving 10 grams sucrose, 20 mg potassium nitrate, 30-gram magnesium sulphate and 10-gram boric acid in 100 ml of distilled water.
- Using a glass rod, stir the solution to mix it well.
- Using a dropper, take some medium and put two drops of it on a clean glass slide.
- Now, take a fresh mature flower and dust a few pollen grains from its stamen on to the drop on the slide.
- After few minutes, place the glass slide on the stage of the compound microscope.
- Observe the slide carefully under a microscope at regular intervals.

### **Observation:**

- Initially pollen grains are observed as uni-nucleated, which at time of liberation becomes two celled.
- The pollen grains take up the medium and start germinating. At first stage, the tube cell enlarges and emerges out of the germ pores of a pollen grain to form a pollen tube.

- The pollen tube further grows in the same manner as it grows through the style to the ovary of the flower.
- The nucleus of the tube cells and germinative cell passes into it and descent towards the tip of a pollen grains.
- Very soon generative cell divides into two male gametes.
- Each male gametes so formed lenticular to spherical in shape.



**Result:**

Pollen germination was observed on the slide.

**Precautions:**

- Freshly plugged flower should be used.
- Procedure should be performed at room temperature.

## Experiment-2

**Aim:** To study the population density of plants by quadrat method.

**Material/Apparatus Required:**

- Fields
- Nails
- Meter scale
- Paper
- Thread
- Hammer

**Principle:**

**Population:** Population is defined as a group of individuals of the same species which inhabit a particular space at a particular time. The number of individuals in a population never remains constant. It may change (increase or decrease) due to many factors like birth rate, death rate and migration.

**Population density:** It represent the number of individuals of the species in any unit area at specific time. The unit area may be small or large, depending on the size and nature of the plant community under study.

Population density is calculated by counting all the individuals present at a given time in a given space, divided by the number of unit area of space.

It is calculated as follows:

$$\text{Density (D)} = \frac{\text{Total no. of individuals of species in all the sampling unit(s)}}{\text{Total no. of sampling unit studied(Q)}}$$

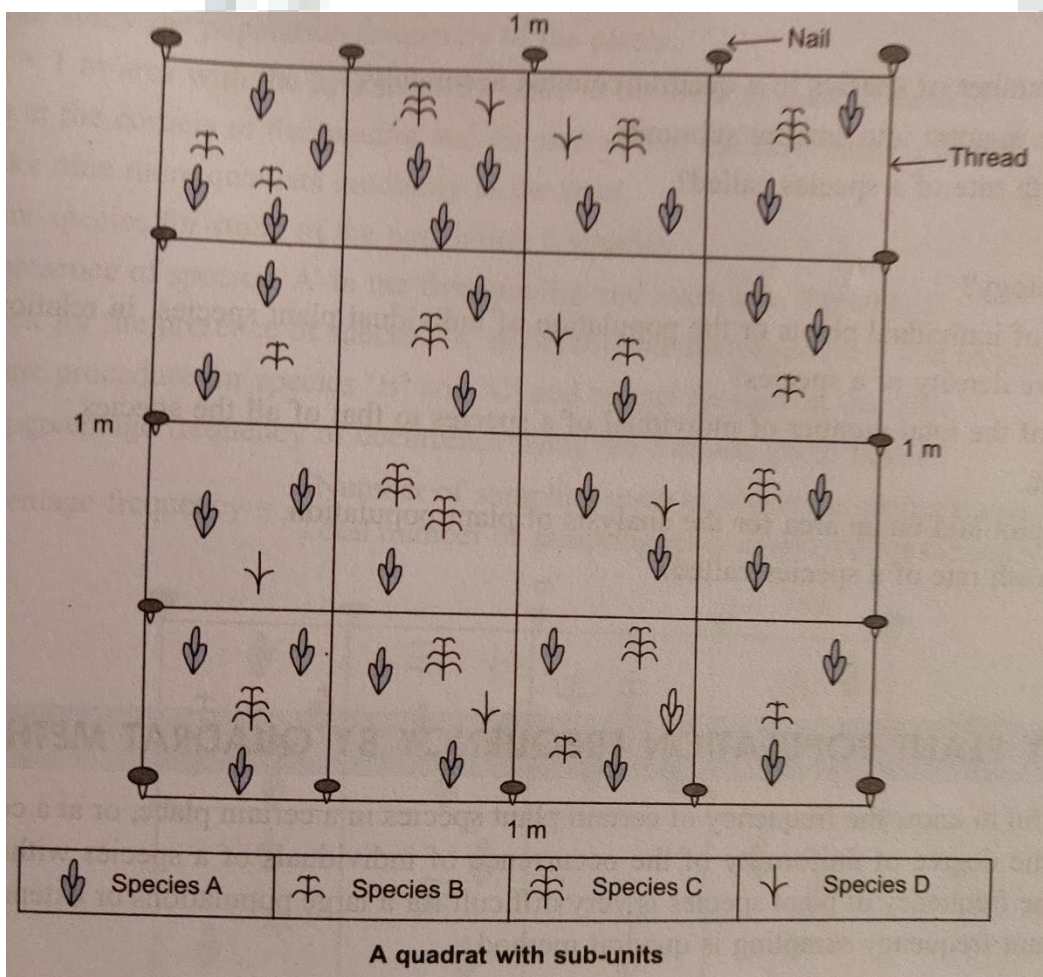
**Quadrat method:** Counting all the individuals in a population is the most accurate method to determine its size. However, this approach is not usually feasible, especially for large population or extensive habitats. Usually, plant population is calculated by quadrat method. A quadrat is a square that encloses an area within a habitat. For herbaceous plants, a meter square (1 m × 1 m) quadrat is normally used.

**Procedure:**

- Select a field to study the population density of the plants.
- Measure 1 meter × 1 meter area with the help of the meter scale in the field to make a quadrat.
- Fix four nails at the corner of the quadrat and tie each end of the nail using a thread.
- Similarly, make nine more quadrats randomly in the field of the study.
- To make the counting easy, divide the quadrat further to make smaller squares.
- Count and mention the number of plants of a particular species in a square and similarly mention the number of plants of another species if present.
- Now, count and add the total number of a particular plant species of all the square to get the total number of a particular plant species of the quadrat.
- Repeat the same experiment for the other quadrats also.
- Record the data in the observation table.

**Observation:**

| Name of the plant species | Number of plants per quadrat |   |   |   |   |   |   |   |   |    | Total number of individuals in all the ten quadrats (S) | Total number of Quadrats (Q) | Population density (D)<br>D=S/Q |
|---------------------------|------------------------------|---|---|---|---|---|---|---|---|----|---|------------------------------|---------------------------------|
|                           | 1                            | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |   |                              |                                 |
|                           |                              |   |   |   |   |   |   |   |   |    |   |                              |                                 |
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|                           |                              |   |   |   |   |   |   |   |   |    |   |                              |                                 |



**Result:**

The population density of a plant species in a given fields can be calculated as:

Population density =  $\frac{\text{Total number of plants in ten quadrats (1 m} \times \text{1 m)}}{10}$

10

**Precautions:**

- Measure the quadrant accurately.
- Mark the quadrant close to each other from one field only
- Do not damage the vegetation while laying the quadrants



## Experiment-3

**Aim:** To study plant frequency by quadrat method

**Material/Apparatus Required:**

- Fields
- Nails
- Meter scale
- Paper
- Thread
- Hammer

**Principle:**

Frequency: Determining plant population frequency using quadrat method has become popular primarily because it is relatively simple and objective. Frequency represents the number of times a plant species is present within a given number of sample quadrants. It is measured by noting the presence of a species in random sample areas which are distributed as widely as possible throughout the area of study. Once analyzed, the sample data enables us to calculate population frequency of the entire population by following method:

$$\% \text{ Frequency} = \frac{\text{Number of sampling units in which the species occurs}}{\text{Total number of sampling units employed for the study}} \times 100$$

Quadrat method: Calculate the plant population frequency using the quadrat is a simple plot of a specific size using for the study of population or a community. It is used in many different scientific disciplines like vegetation assessment, including plant density, plant frequency and plant biomass. Frequency is highly influenced by the size and shape of the quadrats used. The area that is chosen for the study must not be so big that it cannot be sampled adequately, or so small that the habitat is difficult for sampling. For herbaceous plants, a meter square (1 m × 1 m) quadrant is normally used.

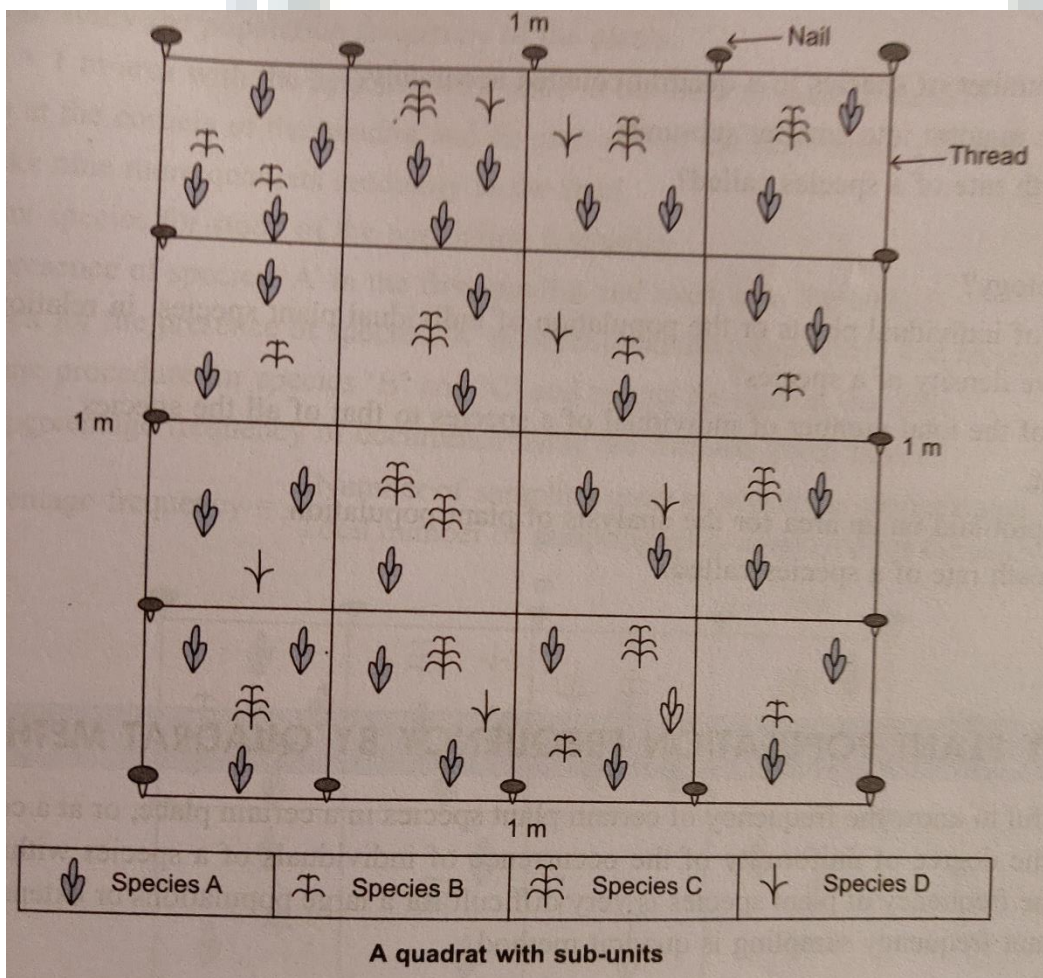
**Procedure:**

- Select a field to study the population frequency of the plants.
- Measure 1 m × 1 m area with the help of meter scale in the field to make a quadrat.
- Fix four nails at the corner of the quadrat and tie each end of the nails using thread.
- Similarly, make nine more quadrats randomly in the fields.
- Select the plant species for the study of population frequency.
- Observe the presence of species 'A' in the first quadrat and mark it in the table
- Similarly, check for the presence of species 'A' in other quadrats respectively and record the data in the table.
- Repeat the same procedures for species 'B' and 'C' and record the data in the table.
- Calculate the percentage frequency of occurrence using the formula given below:

$$\% \text{ Frequency} = \frac{\text{Number of sampling units in which the species occurs}}{\text{Total number of sampling units employed for the study}} \times 100$$

**Observation:**

| Name of the plant species | Number of plants per quadrat |   |   |   |   |   |   |   |   |    | Total number of individuals in all the ten quadrats (S) | Total number of Quadrats (Q) | Population density (D)<br>D=S/Q |  |
|---------------------------|------------------------------|---|---|---|---|---|---|---|---|----|---|------------------------------|---------------------------------|--|
|                           | 1                            | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |   |                              |                                 |  |
|                           |                              |   |   |   |   |   |   |   |   |    |   |                              |                                 |  |
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|                           |                              |   |   |   |   |   |   |   |   |    |   |                              |                                 |  |



**Result:**

Frequency percentage of the species =  $\frac{\text{Total number of quadrats in which species occur}}{\text{Total number of quadrats studied}}$

**Precautions:**

- Measure the quadrant accurately.
- Mark the quadrant close to each other from one field only
- Do not damage the vegetation while laying the quadrants

## Experiment-4

**Aim:** Prepare a temporary mount of onion root tip to study mitosis

### **Material/Apparatus Required:**

- Onion bulbs
- Needles
- Acetocarmine stain
- Glass slide
- Conical flask to germinate onion root tips
- Brush
- Ethanol
- Blotting paper
- Cover slip
- Forceps
- Compound microscope
- Fixative
- Blade
- Dropper
- Corked vial/tube
- Watch glass
- N/10 HCL
- Burner
- Tile

### **Principle:**

A single cell divides to make two cells and these two cells then divide to make four cells, and so on. We call this process “cell division” and “cell reproduction”, because new cells are formed when old cell divides. The ability of cells to divide is unique for living organisms.

Cells divide for many reasons. For example, when the skin of our knee, cells divide to replace old, dead or damaged cells. Cells also divide to help living organism grow. Organisms only grow because cell divides to produce more and more cells. In cell division, the cells that is dividing is called the “parent cells”. The parent cell divides into two “daughter cells”. The process then repeats to form a cell cycle.

Depending on the type of the cell, there are two ways in which cell divide-mitosis and meiosis. Each of these methods of cell division has special characters. One of the key differences in mitosis is a single cell divides into two cells that are replicas of each other and have the same number of chromosomes. This type of cell division is good basic growth, maintenance and repair. In meiosis, a cell divides into cells that have half the number of chromosomes.

**Mitosis cell division:** Mitosis is how somatic cell divides. Somatic cells make up the most of our body tissue and organs including skin, muscles, lungs, gut, and hairs cells. In mitosis, each daughter cells have same chromosomes and DNA as parent cells. The daughter cells from mitosis are called diploid cells having two complete pair of chromosomes. Since the daughter cell have exact copies of their parent cells DNA, no genetic diversity is created through mitosis in normal healthy cells. Mitosis cell division creates two genetically identical daughter diploid cells.



Mitosis cell cycle: Before a cell start dividing, it is in the “Interphase”. It seems that cell must be constantly dividing, but each cell actually spends most of its time in the interphase. Interphase is the period where cell prepare itself to divide and start the cell cycle. During this time, cell gather nutrients and energy. The parent cell also makes a copy of its DNA to share equally between the two daughter cells.

The mitosis division process has several steps or phases of the cell cycle- Interphase, prophase, prometaphase, metaphase, anaphase, telophase, and cytokinesis- to successfully make the new diploid cells. The mitosis cell cycle includes several phases that result in two new diploid cells.

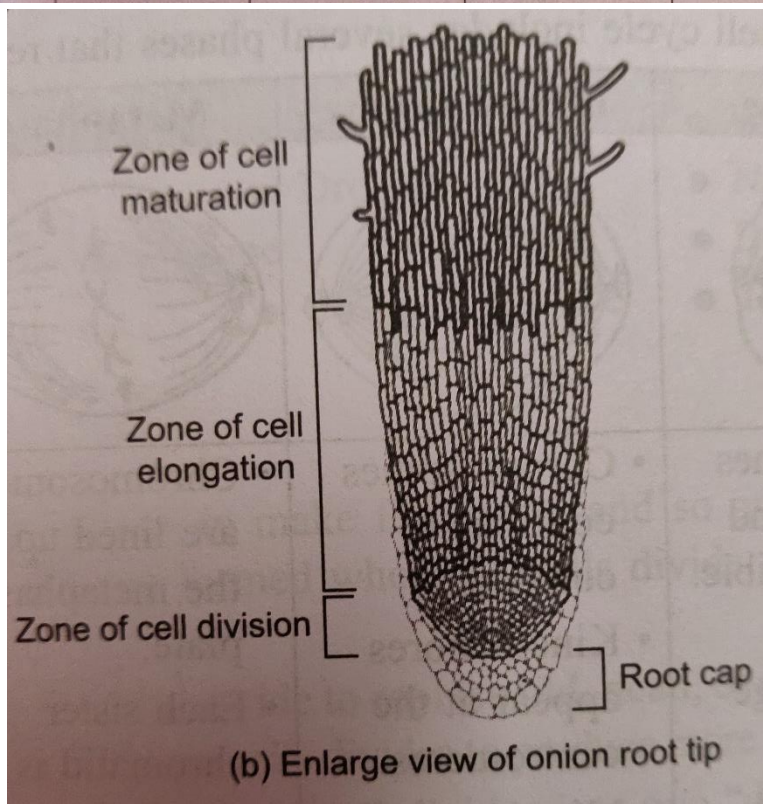
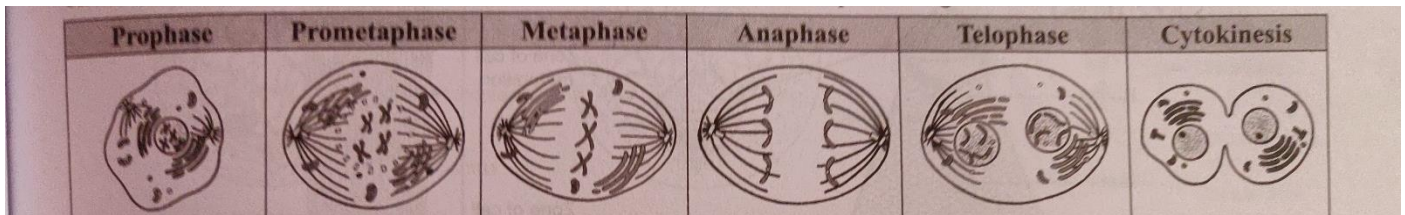
### **Procedure:**

- Take an onion and place it on the tile.
- Carefully remove the dry roots present using sharp blade.
- Grow root tips by placing the bulbs in a conical flask filled with water.
- New roots may take 3-6 day to grow.
- Cut off 2-3 cm of freshly grown roots and let them drop into a water glass.
- Using a forecep, transfer them to vial containing freshly prepared fixative of aceto-alcohol (1: 3: glacial acetic acid: ethanol)
- Keep the root tips in the fixative for 24 hours.
- Using a forecep, take one root and place it on glass slide.
- Using a dropper, place one drop of N/10 HCL on the root tips following 2-3 drops of acetocarmine stain.
- Warm it slightly on burner. Care should be taken that the stain is not dried up.
- Carefully blot the excess stain using blotting paper.
- Using a blade, cut the comparatively more stained portion of the root, retain it on the slide and discard remaining portion.
- After that, put one drop of water on the root tip.
- Mount a coverslip on it.
- Now slowly tap the coverslip slowly so that meristematic tissues of the root tip below the coverslip is properly squashed and spread as a thin layer of cells.
- This prepared slide is now ready for the study of mitosis.
- Place the slide under the compound microscope and observe the different stages of mitosis.
- Various stages of mitosis are prophase, metaphase, anaphase and telophase.

### **Observation:**

- Interphase: The nucleus is metabolically active during this stage and takes part in DNA replication, formation of macromolecules and other energy rich compounds. It is also referred as invisible phase of mitosis since in this stage chromosomes prepare themselves for equitable distribution later on.
- Karyokinesis: It is stage of division which includes following phases: -
  - a. Prophase: Coiling of chromatin occur, nucleolus gradually degenerates from early prophase to late prophase and nuclear envelope start disappearing in late prophase.
  - b. Metaphase: Nuclear membrane disappears completely, chromatids acquire a specific shape and size, moreover chromosomes are attached to poles by spindle fibers through centromere, at equatorial plates.
  - c. Anaphase: Centromere of each chromosome divides into two chromosomes split along longitudinal axis. Two chromatids with its own centromere start moving towards opposite poles. Chromosomes appear V-, L-, J- and I- shaped based upon the position of the centromere.

- d. Telophase: It is reverse prophase. Chromosomes reorganize in nuclei and elongate to form chromatin. Spindle fiber disappears and nuclear envelope reappears.
- Cytokinesis: it is the division of protoplasm following the nuclear division. Normally, it start during middle anaphase and is completed simultaneously with telophase.



**Result:**

All the stages of mitotic cell division are clearly visible in the slide prepared from onion root tips.

**Precautions:**

- The base of the onion should exactly be in contact with water, while growing the roots.
- Filter the acetocarmine stain before use.
- Clean the slide and coverslip thoroughly before use.
- Avoid air bubble in the slide.

## Experiment-5

**Aim:** Isolate DNA from available plant material such as spinach, green pea seeds, papaya, etc.

### **Material/Apparatus Required:**

- 95% ethanol
- Thermometer that will measure 60°C
- 250 mL beaker
- 1000 mL beaker (2)
- Funnel
- Test tube
- Pipets
- Freezer
- Refrigerator
- Hot tap water bath 55-60°C
- Ice water bath 4°C
- Measuring spoons
- Light colored dishwashing liquid/shampoo
- Table salt (1 tsp)
- Distilled water (90 ml)
- Spoon for mixing
- Large onions
- Sharp knife
- Filter paper

### **Principle:**

Deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) are two types of nucleic acid found in living organism DNA act as the genetic material in most of the organisms. Nucleic acid has 3 major components:

- a) Nitrogen base: Purines (Adenine and Guanine) and Pyrimidines: (Cytosine, Thymine and Uracil)
- b) Pentose sugar: Ribose (RNA) and Deoxyribose (DNA)
- c) Phosphate group: nitrogen bases are linked to pentose sugar through N-glycosidic linkage. Two nucleotides are linked through 3'-5' phosphodiester linkage to form dinucleotide.

DNA is made up of two polynucleotide chain, where the backbone is constituted by sugar-phosphate, and the bases project inside. Both polynucleotide chain has anti-parallel polarity.

The process of isolating/ extracting DNA from a cell is the first step for many laboratory procedures in biotechnology. We must be able to separate DNA from unwanted substances of cell gently enough so that DNA is not broken up.

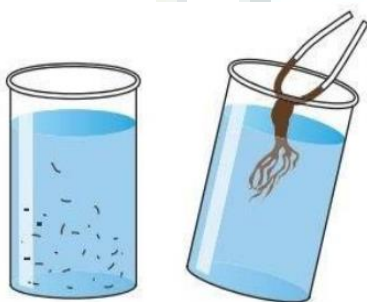
### **Procedure:**

- Put 95% ethanol in the freezer to chill it.
- Prepare two water bath – one at 55-60°C and another filled with ice water around 4°C. For the hot water bath, use large metal pot can be used along with a thermometer with an appropriate temperature range. For ice bath, a mixing bowl filled with ice and water works well.

- In the 250mL beaker, make a solution consisting of 10mL liquid dishwashing detergent/shampoo and  $\frac{1}{4}$  (1.5 gm) of table salt.
- Add distilled water to the beaker to make a final volume of 100mL. dissolve the salt by stirring. This is solution A.
  - a) The liquid detergent causes the cells membrane to break down and dissolve protein and lipid of the cells by disrupting the bond that hold the cell membrane together. The detergent cause lipid and protein to precipitate out. The table salt (NaCl) enables nucleic acid to precipitate out of the ethanol solution because it shields the negative phosphate end of the DNA, causing the DNA strand to come close together.
- Put the 1000 mL beaker with chopped Plant tissue and solution in hot water bath at 55-60°C for 10-12 minutes.
  - a) Do not keep the mixture in hot water bath for more than 15 minutes because the DNA will begin to break down.
  - b) The heat treatment softens the phospholipids in the cell membrane and denature the DNase enzyme which if present, would cut the DNA into small fragment so that it could not be extracted.
- Cool the mixture in an ice water bath, at about 4°C, as shown in figure, for 5 minutes. During this time, smash the tissue with spoon or available apparatus. This step slows down the breakdown of DNA.
- Filter the mixture through a filter paper placed in a funnel over a new 1000 ml beaker. While filtering the tissue, try to keep foam away from getting into the filtrate.
- Dispense the onion solution into fresh test tube with the volume not more than 5 ml.
- Take the cold 95% ethanol and add it to the test tube to create an ethanol layer on the top of about 1 cm.
- DNA is not soluble in layer. When ethanol is added to the mixture, all the components of the mixture, except for DNA, stay in solution while the DNA precipitates out into the ethanol layer.
- Let the solution stand for 2-3 minutes without disturbing it. it is important not to shake the test tube. Watch the white DNA precipitate out into the ethanol layer. When good results are obtained, there will be enough DNA to spool on to a glass rod. DNA has the appearance of white mucus.

**Observation:**

The white DNA precipitate out into the ethanol layer, while the components of the mixture, except for DNA, stay in the solution.



**Result:**

The DNA has been isolated and it appears as transparent, slimy, and white mucus.

**Precautions:**

- Fresh plant materials (onion) should be used for extracting DNA.
- Beakers, test tube and cups must be thoroughly cleaned and dried.
- Handle ethanol carefully because it is highly flammable.





## Experiment-6

**Aim:** Flowers adapted to pollination by different agencies (wind, insects, birds)

### **Theory:**

1. The process of transfer of pollen grains from anther to the stigma of either the same flower or another flower of the same species is called pollination.
2. Pollen grains are immobile, hence require external agencies to reach to the stigma.
3. The external agencies involved in pollination may be abiotic (wind and water) or biotic (insects, birds and snail)

### **Material/Apparatus Required:**

- Fresh flowers
- Magnifying glass
- Glass slide

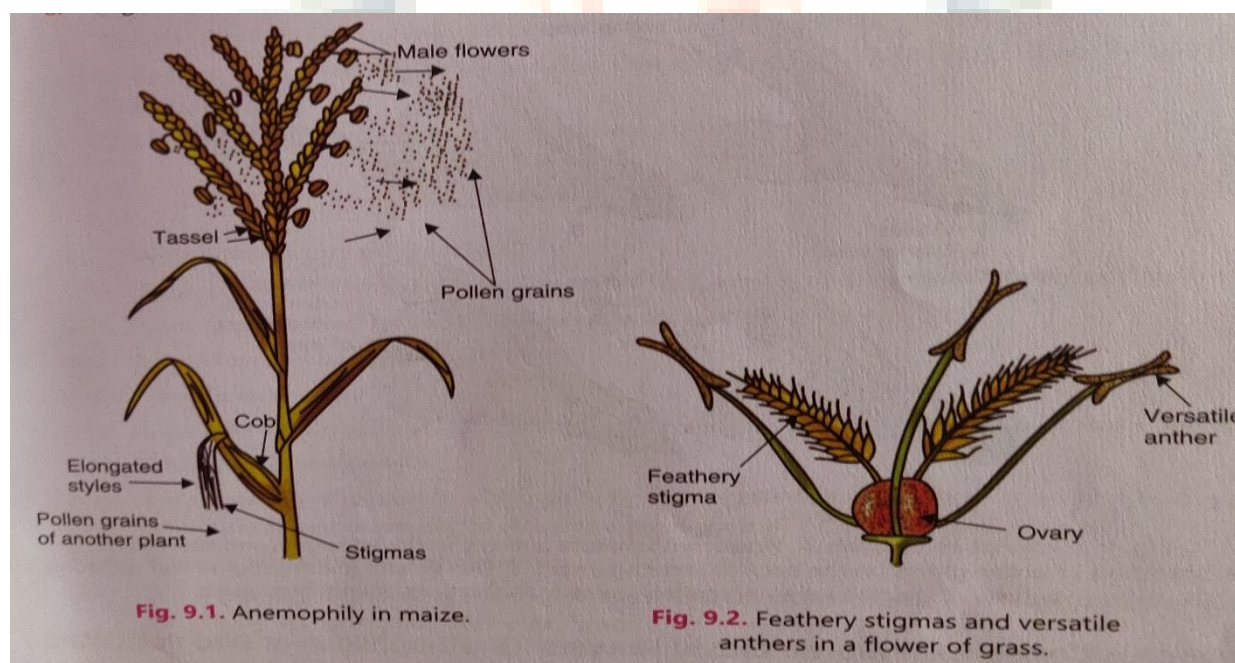
### **Procedure:**

Place the given slide on a slide and observe it with the help of hand lens. Note down the adaptation of the flowers meant for pollination by the external agencies.

### **Maize Flower (Anemophilous or wind pollination flowers)**

The flower of maize shows following adaptations for pollination by wind.

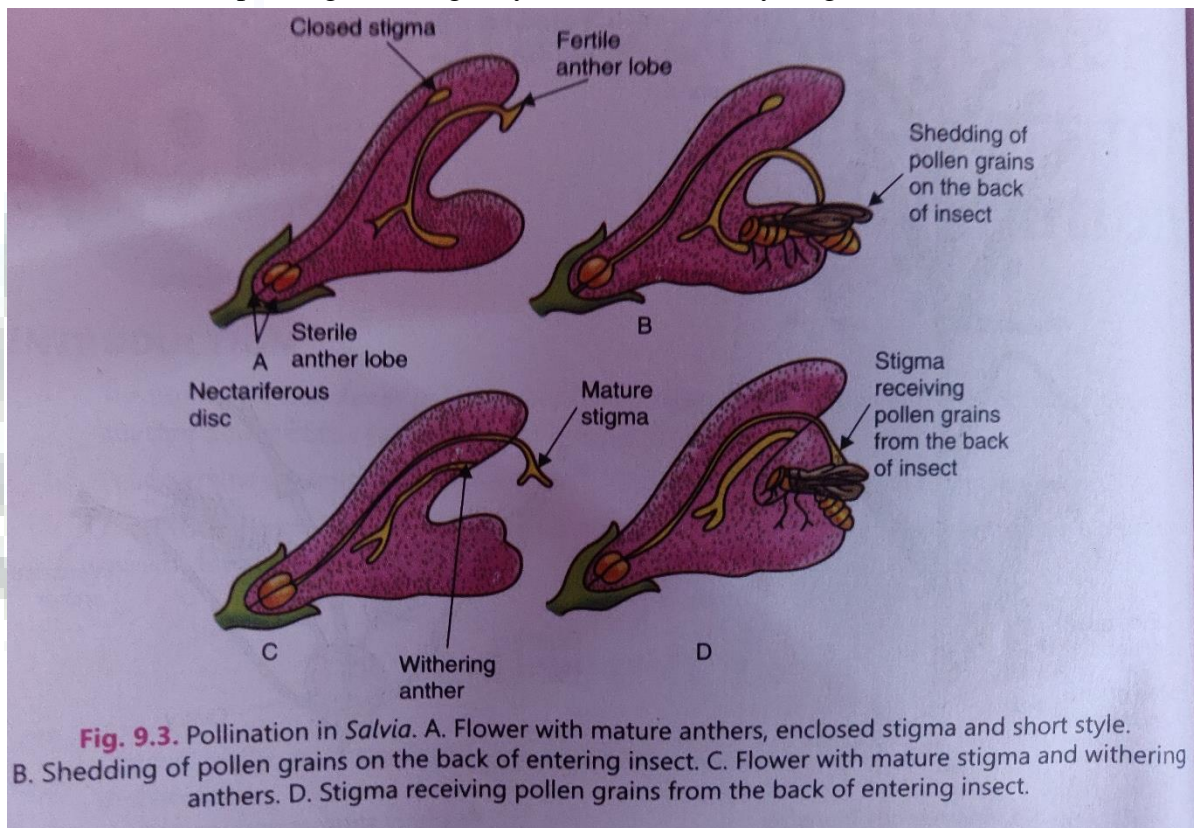
1. The maize plant is monoecious and bears unisexual flowers. The male flowers are born in terminal inflorescence while the female flowers are born in axillary inflorescence.
2. Flower is small, colorless, odourless and nectarless.
3. Flowers are placed in hanging position.
4. Anther and stigma both are exerted (i.e., hang outside the perianth).
5. Anther are versatile, and pollen grains are light, small and dusty.
6. The pollen grains are produced in very large numbers.



### **Salvia Flowers (Entomophilous or Insect pollinated flowers)**

The flowers of *salvia* show following adaptation for pollination by insects.

1. The flower is showy or brightly colored for attracting pollinating insects.
2. Flowers secrete nectar to feed visiting insects. Nectar glands are placed in such position that an insect must touch both anther and stigmas.
3. The flower has landing platform for the insects.
4. Flowers have distinct smell to attract the insects.
5. As the insect move inward a young flower in search of nectar, its head pushes, the anther plate and force the sterile anther lobe to strike against its back.
6. In older flowers the style brings the stigma in such position that it brushes against the back of the insect and collect pollen gains bought by the insect from a young flower.



### **Bignonia/ Callistemon (bottle brush) flowers (Ornithophilous or bird pollinated flowers)**

The flowers of *Bignonia* show following adaptation for pollination by birds.

1. The flowers are usually brightly coloured-red, orange, yellow or blue.
2. The floral parts are commonly leathery.
3. In some cases, the corolla is leathery.
4. The flowers secrete abundant watery nectar or have edible parts.
5. The nectar is secreted in such abundance that drop of it can be brought down by shaking branches.

6. The flowers are generally odorless or without fragrance.





## Experiment-7

**Aim:** Identification of stages of gamete development, i.e., T.S. of testis and T.S. of ovary through permanent slides (from grasshopper/mice)

### **Theory:**

1. Gametes are the sex cells involved in the process of sexual reproduction.
2. The gametes differ from all other cells (somatic cells) of our body in that their nuclei contain only half the number of chromosomes found in their nuclei of somatic cells.
3. Gametogenesis for the formation of sperm is termed as spermatogenesis, while that of ova is called oogenesis.
4. Spermatogenesis occurs in the seminiferous tubules of the testes, whereas oogenesis occurs in ovaries.

### **Material/Apparatus Required:**

- Permanent slides of TS of testes and ovary
- Microscope

### **Procedure:**

- Fix the permanent slide under the microscope. First observe it under the low power and then under high power

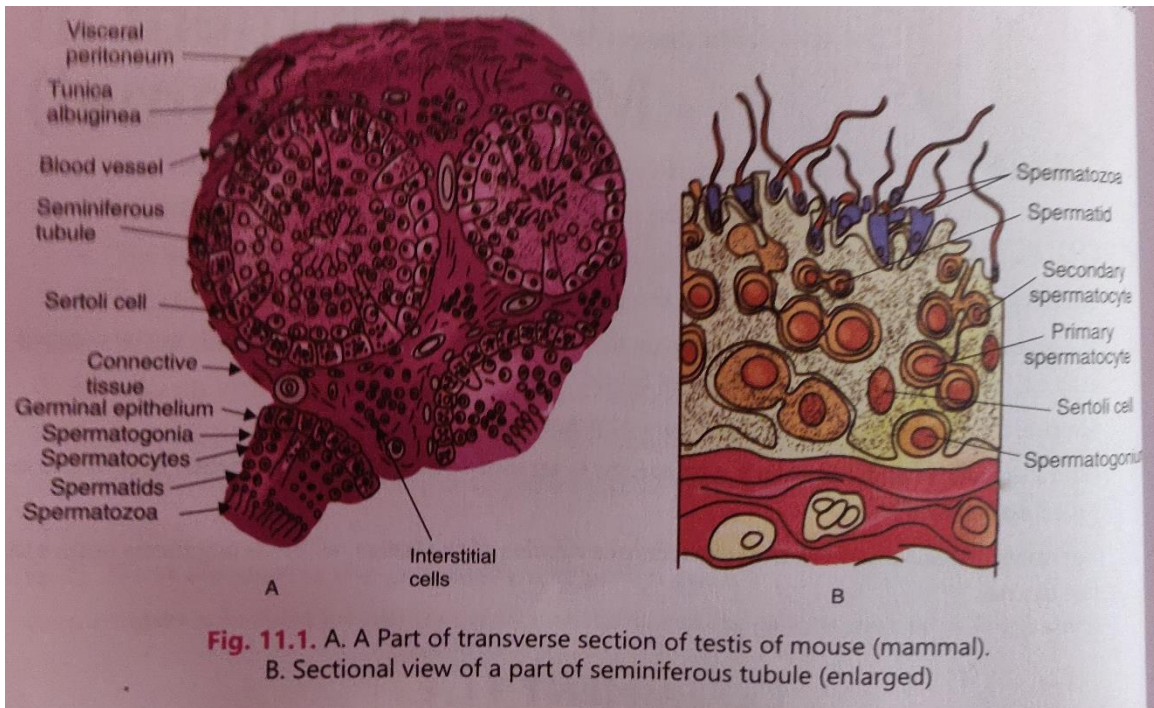
### **Observation:**

#### **T.S of testis**

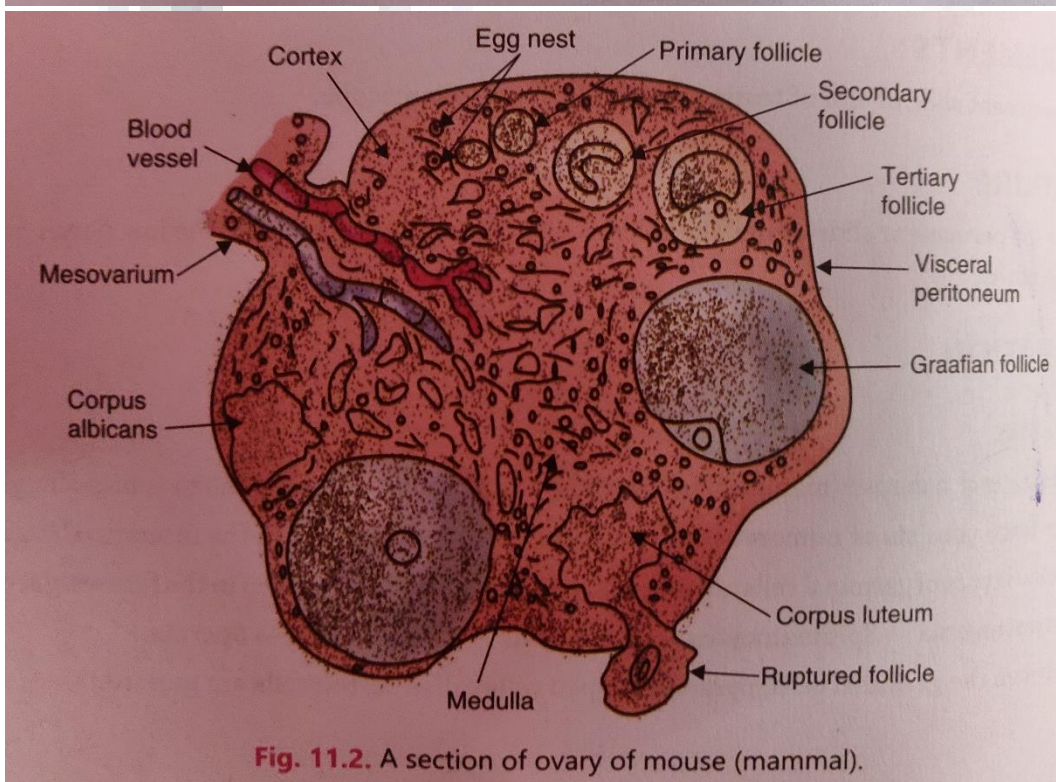
1. The testis of a mouse (mammal) is covered by thick fibrous tissue called tunica albuginea.
2. The testis consists of numerous seminiferous tubules embedded in the interstitial tissue.
3. Various types of germinal cells are present from outside toward lumen in the following sequence. Spermatogonia → spermatocytes → spermatids → spermatozoa → sperms.
4. Between the germinal cells, pyramid shaped cells called Sertoli cells are present.
5. A large number of spermatozoa with their heads embedded in Sertoli cells are present in the lumen of seminiferous tubule.
6. The interstitial tissue also contains Leydig cells, which produce male sex hormones testosterone

#### **T.S of ovary**

1. A mouse ovary is a solid structure bounded by germinal epithelium followed by a thick layer of fibrous tissue, the tunica albuginea.
2. The ovary consists of outer *cortex* and inner *medulla*.
3. The medulla contains many round or oval bodies called *ovarian* or *Graafian follicles* at various stages of development.
4. The medulla also contains blood vessels, nerves fibers and some smooth muscles.
5. Each follicle contains a large ovum surrounded by many layers of *follicle cells*.
6. The cortex contains young and mature follicles.
7. The cortex may also contain a large mass of yellow cells termed *corpus luteum*, formed in an empty graafian follicle after the release of its ovum.



**Fig. 11.1.** A. A Part of transverse section of testis of mouse (mammal).  
B. Sectional view of a part of seminiferous tubule (enlarged)



**Fig. 11.2.** A section of ovary of mouse (mammal).

**Precautions:**

1. First observe the slide under low power and then under high power of the microscope.
2. Use fine adjustment of the microscope for focusing the slide under high power.



## Experiment-8

**Aim:** Meiosis in onion bud cell or grasshopper testis through permanent slides

### **Theory:**

1. Meiosis is a double division which occurs in diploid cells and give rise to four haploid cells each having half the number of chromosomes as compared to the parent cells.
2. It occurs in cells that produce gametes in sexually reproducing organisms.
3. The male gametes (sperms) are produced in testes and the female gametes (egg's) are produced in the ovaries through meiosis.
4. Meiosis comprises of two division- meiosis I and meiosis II. Meiosis I result in reduction of chromosomes number into half. i.e from  $2n$  to  $n$ . hence is called *reductional division*. Meiosis II maintains same number of chromosomes, hence is called *equational division*.

### **Material/Apparatus Required:**

- Permanents slides of different stages of meiosis
- Microscope

### **Procedure:**

- Fix the permanent slide under the microscope.
- First observe it under the low power and then under high power of the microscope.

### **Observation:**

A. Meiosis I (reductional division or heterotypic)

i. Prophase I

- a. Leptotene: appearance of beaded chromosomes.
- b. Zygotene: pairing of homologous chromosomes.
- c. Pachytene: formation of tetrad, crossing over and chiasmata formation.
- d. Diplotene: Two chromosomes of each bivalent move away and chiasmata disappears.
- e. Diakinesis: nucleolus and nuclear envelope disappear and spindle begin to formed.

ii. Metaphase I: Formation of spindle and arrangement of chromosome at equator.

iii. Anaphase I: Separation of homologous chromosome and their movement towards 2 opposite poles of the spindle.

iv. Telophase I: Formation of two haploid nuclei.

B. Meiosis II (homotypic or equational division)

i. Prophase II: Sister chromosome of each chromosome begins to condense.

ii. Metaphase II: Formation of spindle, arrangement of chromosome at equator.

iii. Anaphase II: Movement of daughter chromosomes towards pole of spindle.

iv. Telophase II: Formation of two daughter nuclei from each daughter cell formed by meiosis I.

C. Cytokinesis: division of cytoplasm resulting into four haploid daughter cells form a diploid cell.

### **Precautions:**

1. First observe the slide under low power and then under high power of the microscope.
2. Use fine adjustment of the microscope for focusing the slide under high power.

## Experiment-9

**Aim:** T.S. of blastula through permanent slides (Mammalian)

### **Theory:**

1. Soon after fertilization the zygote undergoes repeated division called cleavage.
2. Cleavage includes a series of successive and rapid mitotic division with transform single celled zygote into multicellular structure called blastula (blastocyst).
3. Cleavage occurs in upper portion of the fallopian tube. It results into a solid mass of cells called morula.
4. At the next stage of development which produces an embryo with about sixty four cells called blastula or blastodermic vesicle.

### **Material/Apparatus Required:**

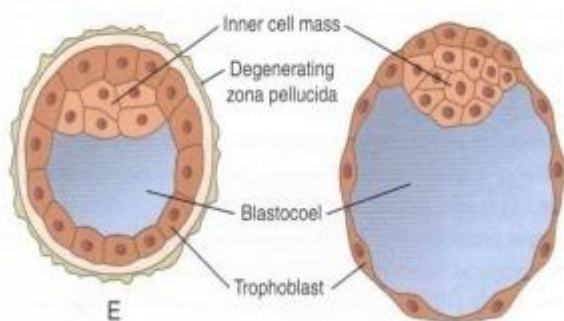
- Permanent slide of blastula and microscope

### **Procedure:**

Fix the slide of T.S of blastula under microscope. First observe the slide under low power and then under high power of the microscope.

### **Observation:**

1. It is a spherical mass of about sixty-four cells.
2. It is composed of an outer envelope of cells, the trophoblast and inner cell mass (embryoblast).
3. Within the envelope there is a fluid filled cavity called blastocoel.
4. The side of the blastocyst to which the inner cell mass is attached is called the embryonic or animal pole, while the opposite side is the abembryonic pole.
5. The inner cell mass is the precursor of the embryo.



### **Precautions:**

1. First focus the slide under low power and then under the high power of the microscope.
2. Use fine adjustment while focusing the slide under high power of microscope.

## Experiment-10

**Aim:** Mendelian inheritance using seeds of different colour/sizes of any plant

### **Theory:**

Mendel hybridized or crossed pollinated plants with alternate forms of one or more traits. He performed monohybrid, dihybrid and polyhybrid crosses and formulated basic law of heredity. These laws are:

1. Law of dominance: when two factors (genes) of a pair of contrasting characters are brought together in a cross, only one of them express itself in the resulting hybrids. The character expressed is said to be dominant and the other which remains suppressive is called recessive.
2. Law of segregation: two factors (alleles) of charters separate or segregate from each other during gamete formation and the paired condition is restored by the random fusion of gametes during fertilization.
3. Law of independent assortment: when the factors (genes) for different characters inherited from parents, do not remain linked in the offspring, but their distribution in the gametes and in the progeny of subsequent generation is independent of each other.

The characters which follow the Mendel laws during inheritance are called mendelian traits.

### **Material/Apparatus Required:**

- Peas seeds samples
- Petri dish
- Notebook
- Pencil/ pen

### **Procedure:**

- Take about 100 pea seeds in a tray.
- Separate out round and wrinkled and put in separate Petri dish
- Note down the number of round and wrinkled seed and calculate their appropriate ratio.
- Repeat the process for the other contrasting traits of the seed i.e., yellow and green colour.

### **Observation:**

| Sr. no. | Character / traits of the seed | Total no. of seed observed | No. of seed showing contrasting form of the trait | Appropriate ratio |
|---------|--------------------------------|----------------------------|---|-------------------|
| 1       | Seed shape (Round/ wrinkled)   |                            |   |                   |
| 2       | Seed shape (yellow/green)      |                            |   |                   |

### **Result:**

The contrasting forms in both the traits of pea seed (i.e., seed shape and seed colour) show an approximate ratio of \_\_\_\_\_.

### **Precautions:**

Observe the contrasting traits carefully and take sufficient large number of seed for analysis to minimize the error.

## Experiment-11

**Aim:** Prepared pedigree charts of any one of the genetic traits such as rolling of tongue, blood groups, ear lobes, widow's peak and colour blindness

### **Theory:**

1. A record of inheritance of certain genetic traits for two or more generations presented in the form of a diagram or family tree is called as pedigree
2. In pedigree, a square represents a male, a circle represents a female, solid symbol represents traits under study and a cross or shade (of any type) in the symbol signifies the carrier of recessive allele.
3. Parents are shown by horizontal line while their offspring are connected to it by vertical line.

**Autosomal Dominant Trait-** Blood Groups, Free hanging earlobes, Widow's Peak, Rolling of tongue. The encoding gene for these genes is present on any of the autosomes. In these traits, the mutant allele is dominant.

Such type of traits exhibits the following features:

- a) The traits get transmitted from the parents to either gender.
- b) It affects males and females equally.
- c) The trait is present in each of the generations, i.e., the pedigree is vertical.
- d) Some common traits of this type include blood groups, polydactyly, brachydactyly, the dimple in cheeks, etc.

### **Autosomal Recessive Trait**

The mutant allele of such traits is recessive.

Salient features of such type of traits include:

1. It is found equally in multiple male and female siblings whose parents are carriers.
2. Homozygous siblings for defective alleles, but parents are heterozygous.
3. If men and women who are genetically related are married to each other, they might exhibit this trait.

### **X-Linked Dominant Traits**

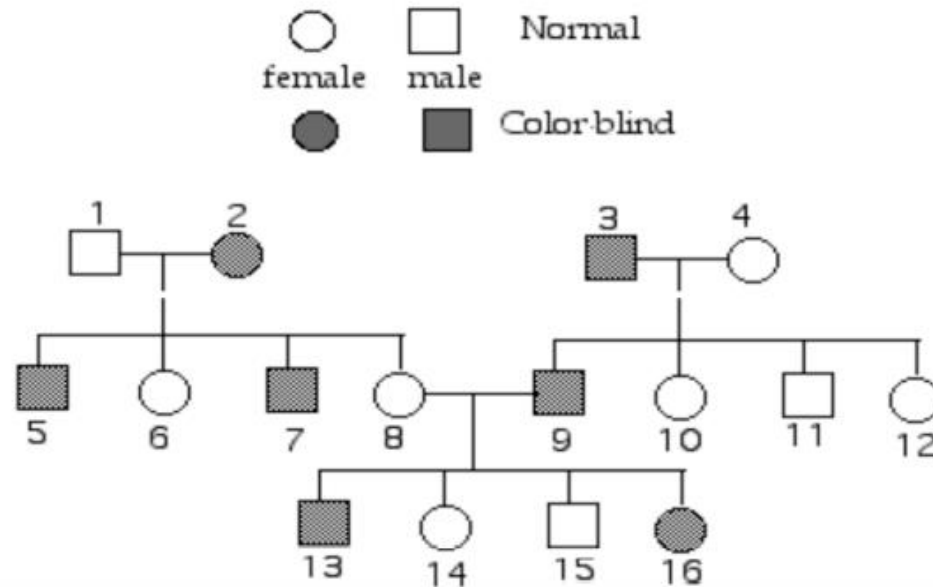
The encoding gene for such traits is located on the X chromosome. The mutant allele is dominant in this trait.

The features of such type of traits are:

1. Inheritance is vertical and is found in all the generations.
2. If the female is affected, half of her sons are also affected.
3. If the male is affected, all the daughters will be affected but no sons will be affected, i.e., there is no male-to-male transmission

## X-Linked Recessive Traits- Colour Blindness

In humans, color blindness is caused by a **recessive sex-linked** allele. Examine the pedigree of color-blindness below. On the diagram, label the genotypes of every individual. Note: If the gene is on the Y chromosome (Y-linked), we would write it as  $Y^b$ , and a male with this trait would be written as  $XY^b$ . If the allele is on the X chromosome (X-linked), we would write it as  $X^b$ , and a heterozygous female would be  $X^B X^b$  (1 pt)



In such type of traits, the mutant allele is recessive to the wild type allele. The features of X-linked recessive traits include:

1. This is expressed only by homozygous females but homozygous and hemizygous males.
2. If the female is the carrier, about half the sons are affected. If the female is homozygous, 50% of the daughters and 100% of the sons can be affected. That is why the male population is the most affected.

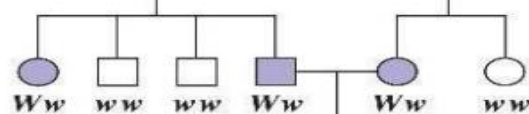
## Y-chromosome Linked Traits

The gene for such traits is present on the Y-chromosome. Any trait linked to Y-chromosome is found only in males and not in females because the Y-chromosome is present only in males. All the sons of the affected male exhibit the trait, whereas, none of the daughters exhibits the trait.

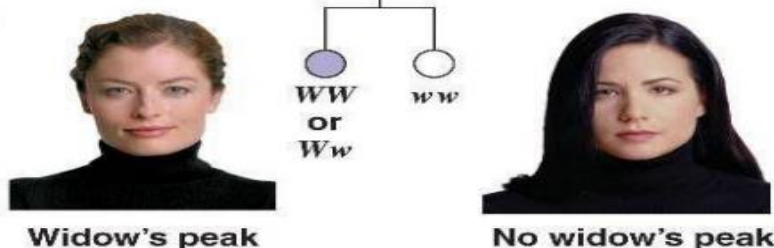
1st generation (grandparents)



2nd generation (parents, aunts, and uncles)



3rd generation (two sisters)





**Material/Apparatus Required:**

- Information about traits in a family for more than one generation

**Procedure**

1. Select a family with anyone of the monogenic traits like rolling of tongue, blood groups, ear lobes, widow's peak, and colour blindness.
2. Ask the person exhibiting the trait as to who in his/ her family has the trait in question.
3. Prepare a pedigree chart on the basis of the information collected, using appropriate symbols.
4. Examine the pedigree chart carefully to find out whether the disease is autosomal recessive, autosomal dominant, X-linked dominant or recessive, and Y-linked dominant or recessive.

**Results:**

Pedigree charts explains the genetic traits.



## Experiment-12

**Aim:** Controlled pollination - emasculation, tagging and bagging

### **Theory:**

1. Plant breeding is a science of changing and improving the heredity of plant and production of new varieties which are far better than original type.
2. Hybridization is a method by which improved varieties of economical important plants/crops are produced by crossing two or more genetically different plants.

### **A. Emasculation**

**Material/Apparatus Required:** forceps or scissors method of emasculation.

#### **Comment**

- i. This method is employed in the crops having flower sufficiently large size.
- ii. The instrument used in this method includes pocket lens, forceps, needle, scissors, scalpel, hair brush, etc.
- iii. In this process anther are removed from flowers before their maturation.
- iv. The anther are cut with the help of sterilized forceps's or scissors.

**Material/Apparatus Required:** Hot or cold water and alcohol emasculation

#### **Comment**

- i. This method of emasculation is employed the crop having small flowers like paddy, sorghum, etc.
- ii. In this method the penicles (clusters of flowers are dipped in hot water (44 - 45°C) for 1 -10 minutes to kill the anthers.
- iii. In the same way emasculation is done with cold water or alcohol.

### **B. Tagging and Bagging**

**Material/Apparatus Required:** Bagging, tagging and labelling.

#### **Comment**

- i. After emasculation, the flower are covered with small bags to prevent pollination with undesired pollen grains.
- ii. These bags are made up of polyethene, paper, muslin cloth or parchment paper.
- iii. The bags are punctured or made perforation so as to provide aeration to the flowers.
- iv. The flower of male parents are also protected in bags to prevent mixing of their pollen grains with foreign pollens.
- v. After dusting of the desired pollen grains on the emasculated flowers, the bags are re-tagged.
- vi. A label of paper is tagged on the plant which display the date of emasculation, crossing and brief account of the parent.

## Experiment-13

**Aim:** Common disease causing organisms like Ascaris, Entamoeba, Plasmodium, any fungus causing ringworm through permanent slides, models or virtual images or specimens. Comment on symptoms of diseases that they cause

### **Material/Apparatus Required:**

Preserved slides or specimens of disease-causing organisms like Ascaris, Entamoeba, Plasmodium and Ringworm

### **Procedure**

Observe the specimens or slides and identify the organism on the basis of its features.

### **Observation:**

#### **A. Ascaris**

Phylum: Aschelminthes

Class: Nematoda

Type: Ascaris lumbricoides

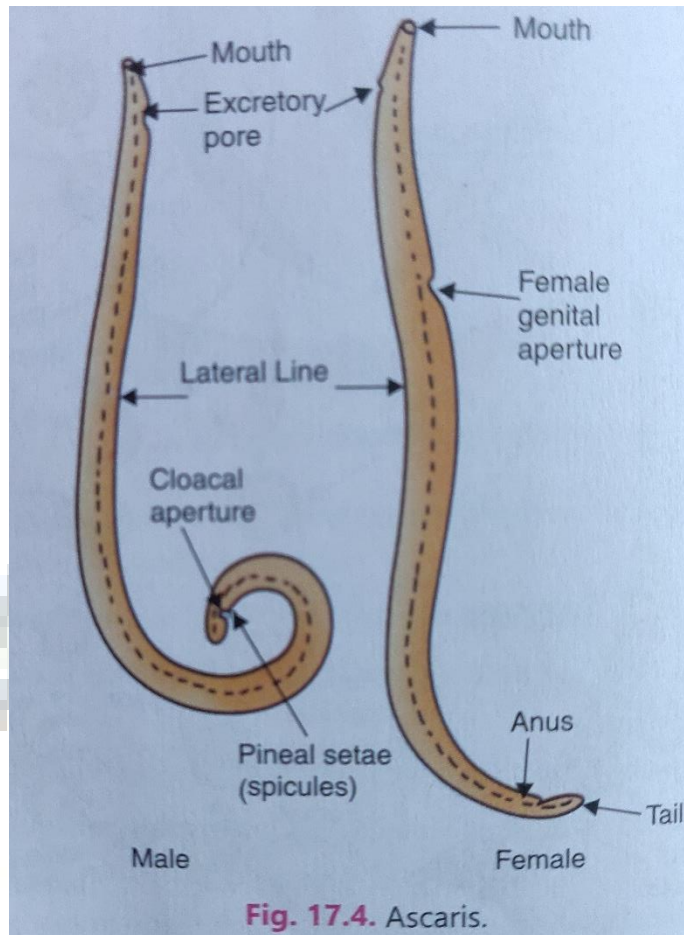
Ascaris exhibits the following characteristic features:

- 1) It has a long, cylindrical and unsegmented body.
- 2) The male and female organisms are separate.
- 3) It bears a mouth at the anterior end surrounded by three lips.
- 4) There is an excretory pore on the ventral surface slightly behind the anterior end.
- 5) A pair of penial spicules are present in the male worms close to the cloacal opening.
- 6) The female genitals are present at about one-third distance from the anterior end.

Ascariasis is the disease caused by Ascaris lumbricoides or roundworm.

Symptoms:

- Abdominal cramping
- Abdominal swelling
- Nausea
- Vomiting
- Fever



## B. Entamoeba

Phylum: Protozoa

Class: Rhizopoda

Type: *Entamoeba histolytica*

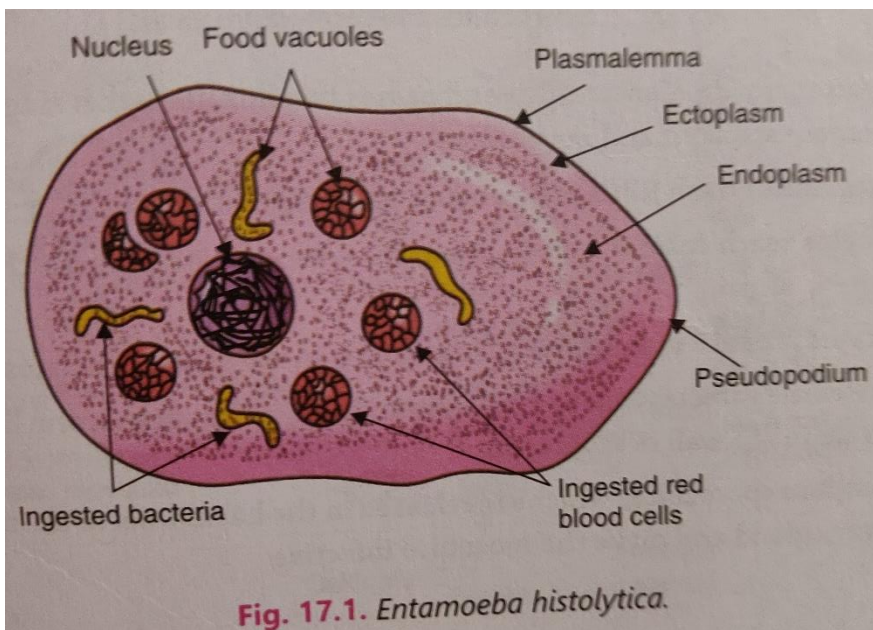
Following are the characteristic features of *Entamoeba*:

- 1) It is a unicellular organism with an irregular shape.
- 2) It consists of a few food vacuoles. The contractile vacuole is absent.
- 3) Cysts with four nuclei are present.
- 4) It consists of a nucleus located eccentrically in the cell.

*Entamoeba histolytica* is an organism found in the intestines of humans that is responsible for causing amoebic dysentery

Symptoms:

- Abdominal pain
- Watery diarrhoea with mucus, blood and pus
- Fatigue
- Fever
- Nausea
- Vomiting



### C. Plasmodium

Phylum: Protozoa

Class: Sporozoa

Type: Plasmodium vivax

Plasmodium can be identified by the following characteristic features:

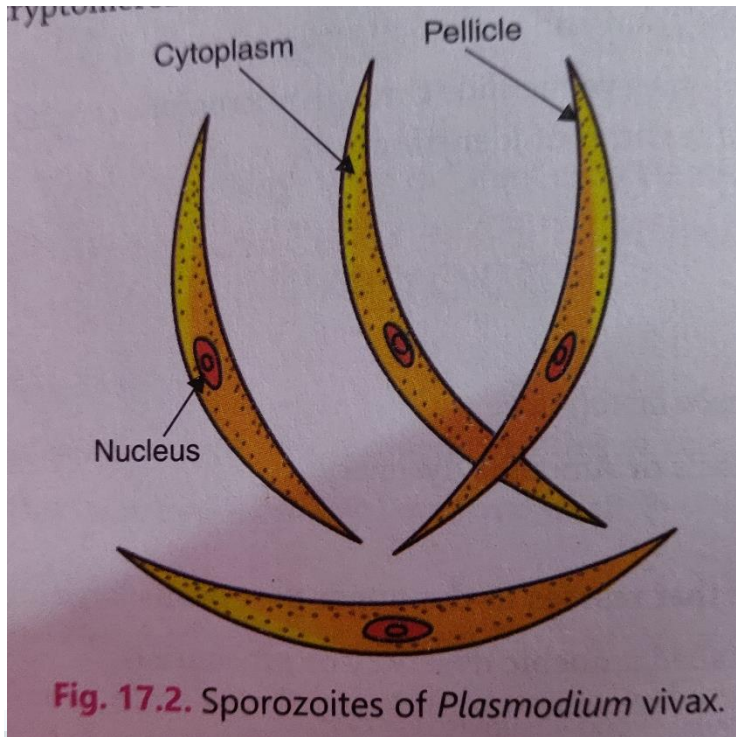
- 1) It is a unicellular endoparasite found within the red blood cells of the diseased person.
- 2) The parasite is mostly diagnosed at the “signet ring” stage where the parasite appears as a round body.
- 3) There is a big vacuole present inside the cell. The cytoplasm is accumulated at one place and contains the nucleus.

*Plasmodium vivax* is a protozoan parasite that causes malaria in humans. The infected female anopheles bites a healthy person and transmits the sporozoite into the peripheral blood vessels of humans, thereby, causing malaria.

Symptoms:

- High fever
- Shaking chills from moderate to severe.
- Headache
- Vomiting
- Nausea





#### D. Ringworm

Kingdom: Fungi

Class: Deuteromycetes

Type: *Trichophyton rubrum*

Trichophyton or ringworm fungus has the following characteristic features:

- 1) This fungus feeds on the keratin of the skin of human beings.
- 2) The hyphae are waxy and can be smooth or cotton-like.
- 3) Hyphae that are not stained are yellowish-brown, reddish-brown or white in colour.

Ringworm is a communicable fungal infection of the skin.

Symptoms:

- Scaly, itchy skin
- Red and raised patches
- They are redder at the periphery than at the center and forms a ring-like appearance

