Exercise 2

Aim: To calculate percentage of pollen germination

Principle: In nature, pollen grains germinate on the compatible stigmas of the carpel. Pollen grains can also be induced to germinate in a synthetic medium. During germination, **intine** (inner wall) of pollen grain emerges out as **pollen tube** through one of the germ pores in **exine** (outer wall).

Requirement: Calcium nitrate, boric acid, sucrose, distilled water, petridish, slides, coverslips, brush, needle, microscope, and mature pollen grains of *Tradescantia*/balsam/Jasmine/lily/pomegranate/grass/*Vinca*/China rose/*Petunia*.

Procedure

(i) Prepare the pollen germination medium by dissolving 10g sucrose, 30mg calcium nitrate and 10mg boric acid in 100ml of distilled water. Alternatively 10% sucrose solution can also be used.

(ii) Take a drop of medium or 10% sucrose solution on a cover slip and sprinkle mature pollen grains on the drop.

(iii) Invert the cover glass on to a slide

(iv) After 10 minutes, observe the slide under microscope.

(v) Count (a) total number of pollen grains seen in the microscope field, and (b) the number of pollen grains that have germinated.

Observation

Several pollen grains germinate and put forth pollen tubes. Count the total number of pollen grains and the number of germinated pollen grains in 3-5 different microscope fields. Tabulate your observations and calculate the percentage of pollen germination.

Name of the plant used as source of pollen……

Number of pollen grains in a field of microscope = N

Number of germinated pollen grains in a field of microscope = n

Percent pollen germination = $\frac{n}{N} \times 100$ or $\frac{100n}{N}$

Number of observation	Total number of pollen (N)	Total number of pollen germinated (n)	% pollen germination $\frac{n}{N} \times 100$
1.			
2.			
3.			
4.			
5.			
Average			

Discussion

Although pollen grains of many species germinate in this medium, the percentage of germinations and the time taken for germination varies in different species. Draw a germinating pollen grain and label.

Questions

1. How many pollen tubes emerge from a pollen grain?
2. What does the pollen tube carry?
3. Can you explain as to why some pollen grains fail to germinate?
4. Why do we use sucrose as the medium for pollen germination?

Exercise 3

Aim: To study pollen tube growth on stigma

Principle: Pollen grains germinate and form pollen tubes after they get deposited by the process of pollination on compatible stigma. Pollen tube, made up of cellulose, is an extension of the inner wall of pollen grain (intine). It emerges through one of the germ pore and passes through tissues of stigma and style to reach the ovule. The growing pollen tube is observed by staining with cotton blue.

Requirement: 5–6 excised styles with stigma of *Petunia*/grass/maize/sunflower/*Abelmoschus* (Lady's finger), beaker, water, slides, cover slips, cotton blue stain, microscope, brush, needle.

Procedure

(i) Place the stigmas in boiling water in a beaker for softening the tissues for 5–10 minutes.

(ii) Stain with cotton blue for 3–5 minutes and wash with water to remove excess stain.

(iii) Mount one stigma in a drop of glycerine on a slide. Place a cover slip on the stigma and gently press the cover slip on the material. Observe the slide under a microscope.

(iv) If you fail to observe pollen tubes mount another stigma.

Observation

Look for long blue-coloured tubular structures traversing through the tissues of stigma and style (Fig. 3.1).

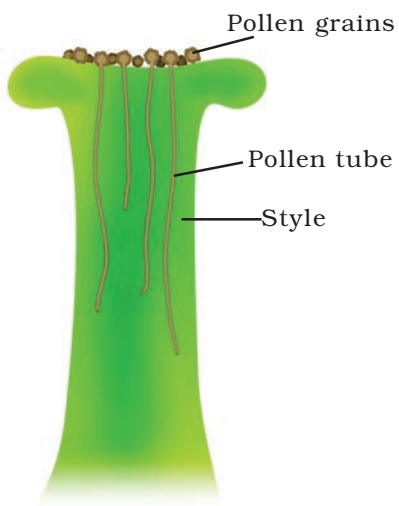

Fig.3.1 Growth of pollen tube in the style of a carpel

Discussion

Pollen tubes are seen amidst the stylar tissue. Many pollen tubes may be seen. Trace the origin of pollen tubes to the pollen grains present around the surface of the stigma.

Questions

1. Can pollen grains of one plant species germinate on stigma of other species? Give reasons.
2. Do all pollen tubes reach the ovules?
3. Are all the pollen tubes of equal length? If not, why?