EXPERIMENT

AIM

To study the pollen germination on stigma through permanent slide.

THEORY

The pollen grains are the male gametophytes in angiosperms. These are produced in anthers as a Result of a process called microsporogenesis. The microspore mother cell in anthers divides meiotically to produce tetrad of pollens, each has haploid number of chromosomes. Each pollen is surrounded by two wall layers outer exine and inner intine.

Exine

It is outer tough protective layer. It has sporopollenin which is the most resistant biological material known so far. At some places exine is either very thin or absent, such regions in exine are called germ pores.

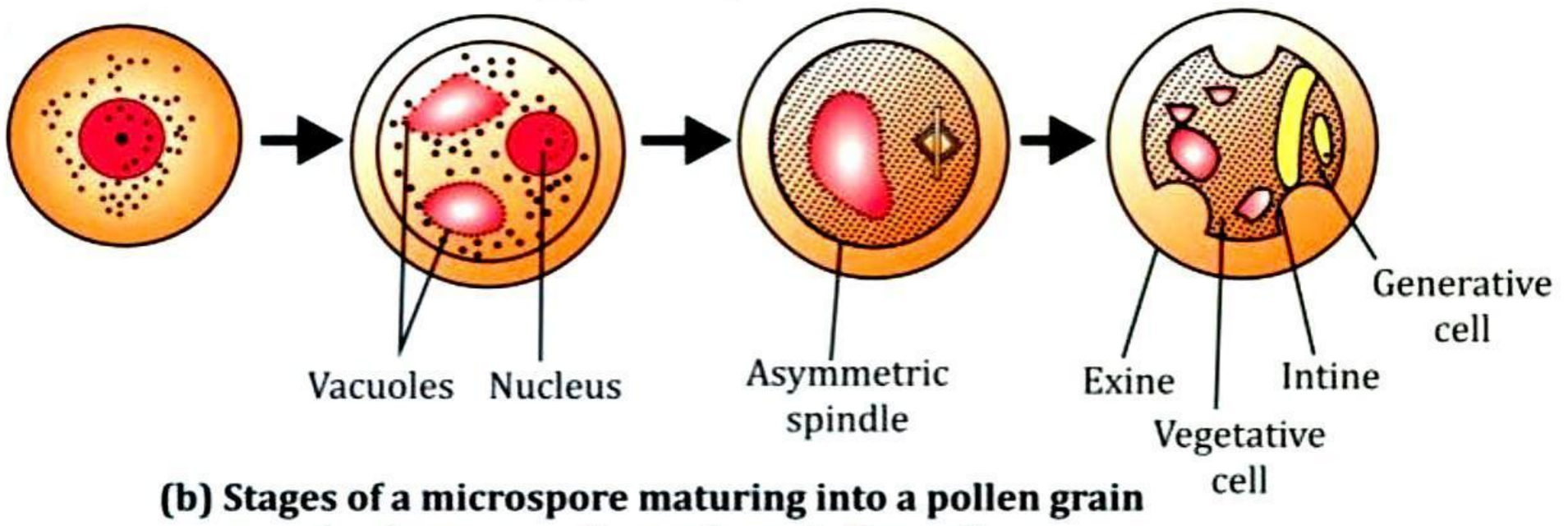
Intine

It is inner smooth layer made up of pectocellulose deposition in its cell wall. Intine gives rise to pollen germ tube through germ pores present in exine. Before the discharge from microsporangium, each pollen grain possesses two cells initially. These are

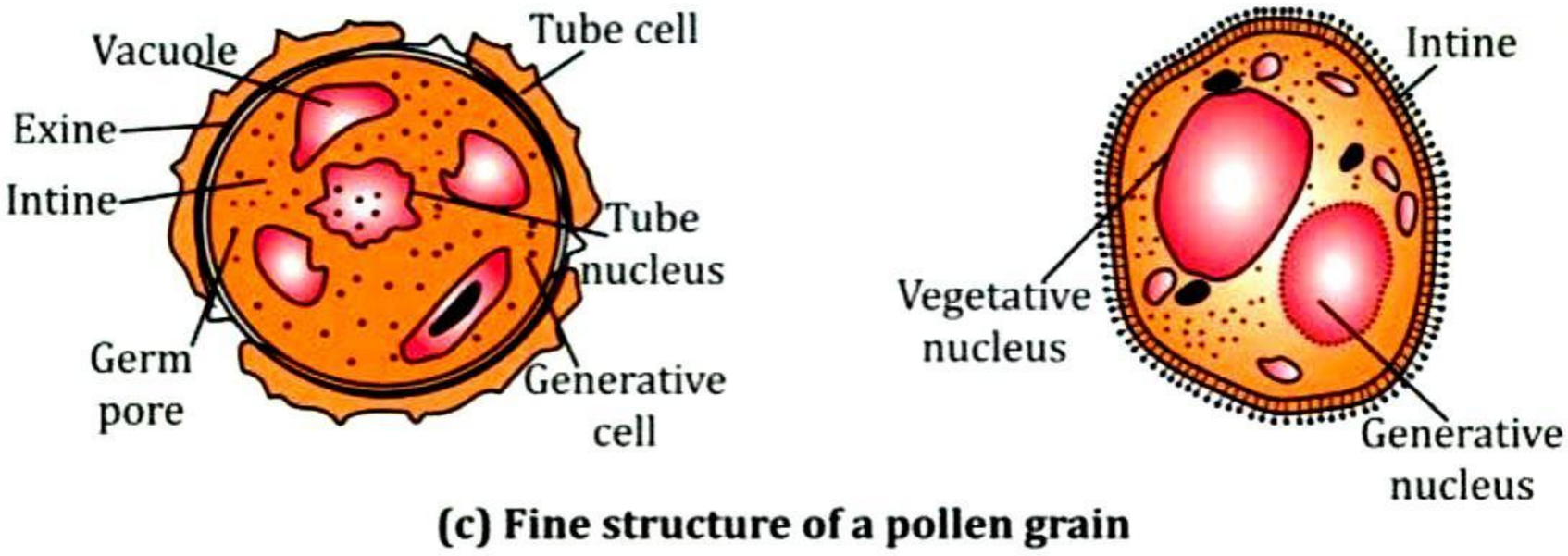
(i) Large vegetative cell

(ii) Small generative cell as shown in figure below:

(a) Pollen grain tetrad



(b) Stages of a microspore maturing into a pollen grain having generative and vegetative cell



(c) Fine structure of a pollen grain

Generative cell of a pollen grain

The generative cell later divides to form two male gametes. At the time of pollen shedding from anthers, it is a 3-celled structure till it reaches stigma by the process of pollination. The pollen germination is initiated by certain chemicals present in the stigma and the growth of the pollen tube (made up of cellulose) is supported by sugary substances called nectar secreted by cells of stigma. The pollen tube emerges through one of the germ pore and passes through tissues of stigma and style to reach the ovule and discharge its male gametes. One male gamete fuses with egg cell and the other male gamete fuses with secondary nucleus of temale gametophyte. This phenomenon is called double fertilisation, a characteristic feature of all angiosperms.

REQUIREMENTS

Pemanent slide of pollen germination with stigma, compound microscope, drawing sheets, pencil. eraser, etc. For temporary slide preparation : 5-6 excised styles with stigma of Petunia/grass/maize/ sunflower, beaker, water, slides, coverslips, cotton blue stain, brush and needle.

PROCEDURE

Using Temporary Slide

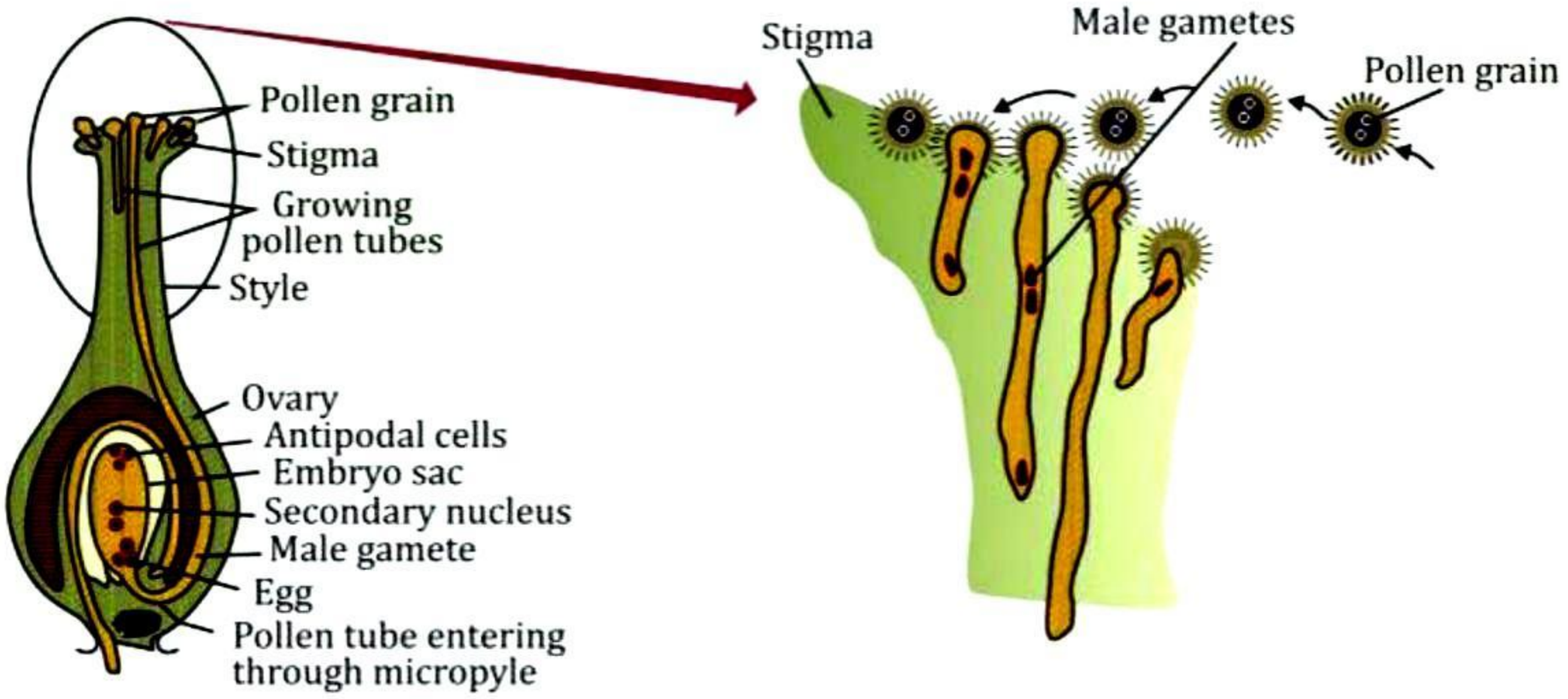
1. Place the freshly excised stigma and styles in boiling water in a beaker for 5-10 minutes. This will soften the tissues and also prevent them from drying.
2. Add cotton blue dye using a dropper onto the tissue placed on a glass slide.
3. Wait for 3-5 minutes and then wash-off the excess stain with water.
4. Mount one stigma in a drop of glycerine on a slide.
5. Place a coverslip on the stigma and gently press the coverslip on the material.
6. Place the temporary slide under a compound microscope and observe initially under low magnification followed by higher magnification.
7. Draw a well-labelled diagram of the structures seen.

Using Permanent Slide

1. Pick up a good permanent slide of pollen germination.
2. Place it under low power of microscope and observe the slide.
3. Focus structures seen in low power, then in high power to have clear view of details of pollen tube.
4. Draw a well-labelled diagram of the structures seen.

OBSERVATIONS

The slide shows the presence of many germinating pollen grains on the stigma of the carpel. The pollen tubes of the germinating pollen grows through the style, transversing the tissues of the style. The slide shows many such growing pollen tubes of varied length.



(a) L.S. of carpel showing stigma, style and overy

(b) Pollen grain germinating on stigma

Pollen grain germination

RESULT

As the pollen tube grows into style, it reaches to the ovule and carries the two male gametes; one to the egg cell and another to secondary nucleus for fertilisation to occur inside the ovule.

PRECAUTION

- The prepared permanent slide should be handled carefully.
- Draw proper figures of the structures as seen under low and higher magnifications of the compound microscope.
- Label the figures accurately.

VIVA VOCE

Q1. What are pollen grains?

Ans. Pollen grains are germinated microspores which represent the male gametophyte.

Q2. Name the cells present in the pollen grain.

Ans. A pollen grain possesses a tube cell (vegetative cell) and a generative cell.

Q3. Which part of pollen forms the pollen tube?

Ans. The intine wall of pollen grain gives rise to pollen tube after germination.

Q4. What are germpores?

Ans. Germpores are the places where exine is either very thin or absent.

Q5. Do all pollen tubes reach to ovules?

Ans. No, all pollen tubes do not reach to the ovule.

Q6. What stimulates the germination of pollen grain?

Ans. Sugary substance called nectar present on the stigma stimulates the germination of pollen grain.

Q7. From where does the pollen tube obtain its nourishment during growth?

Ans. Pollen tube obtains its nourishment from the transmitting tissue of the style during its growth.

Q8. Are all pollen tubes of equal length? If not, why?

Ans. Most of the pollen germinate on stigma, but all are not compatible to grow because there exist genetic incompatibility. Those pollens which are not compatible, their growth is not supported by stylar tissue and they stop growing. Only compatible pollen tubes grow further and reach ovule. That is why all pollen tubes are not of the same length.

Q9. Name the part of gynoecium that determines the compatible nature of pollen.

Ans. Stigmatic tissue determines the compatibility of a pollen.

Q10. Can pollen grains of one plant species germinate on stigma of other species?

Ans. The germination of pollen grain accurs on the stigma of the gynoecium. Most of the time pollen germinate as they get deposited on stigma. Their further growth, i.e. growth of the germ tube depends on the genetic compatibility of the two species. If these are compatible, the germ tube will grow if not, the germ tube will stop growing.

Q11. Name the material present in the exine of pollen which makes it overcome adverse climatic conditions.

Ans. Sporopollenin is present in the exine of pollen. This is the most resistant biological material known so far.

Q12. Mention the role of pollen tube in sexually reproducing flowering plants.

Ans. Role of pollen tubes is to carry 2 male gametes to the female gamete present in the embryo sac of female gametophyte.

Q13. Where does pollen tube arise in a pollen grain at the time of pollen germination?

Ans. The pollen tube takes its origin from intine through germ pore present in exine.