MALE AND FEMALE GAMETOPHYTE DEVELOPMENT

- Specialized cells in the anther undergo meiosis to produce four haploid microspores that develop into pollen grains.

- Similarly, a cell within the ovule divides meiotically to produce four haploid megaspores, one of which survives and undergoes three mitotic divisions to produce the cells of the embryo sac.

- The embryo sac represents the mature female gametophyte.

- The pollen grain, with its germinating pollen tube, is the mature male gametophyte generation.

- The two gametophytic structures produce the gametes (egg and sperm).

- **Plants undergo an alternation of generations life cycle that involves a multicellular haploid generation, called the gametophyte, and a multicellular diploid generation, called the sporophyte.**

- **Sexual reproduction is initiated with sporogenesis, during which specialized cells (mother cells) within the sporophyte undergo meiosis and give rise to haploid spores.**

- **Spores undergo gametogenesis, a process of cell proliferation and differentiation, to develop into multicellular gametophytes, which then produce the gametes (sperm and egg cells).**

- **Fusion of egg and sperm to form the zygote, followed by embryo body plan development gives rise to the sporophyte, thereby completing the life cycle.**

- **Angiosperms, or flowering plants, are heterosporous, producing two types of spores that develop into two types of unisexual gametophytes.**

- **The first spore type is the megaspore.**

- **During megasporogenesis, diploid megaspore mother cells undergo meiosis and give rise to haploid megaspores, which then, during megagametogenesis, develop into haploid female gametophytes.**

- **The second spore type is the microspore.**
- **During microsporogenesis, diploid microspore mother cells give rise to microspores, which then undergo microgametogenesis and develop into male gametophytes**

Male gametophyte development in angiosperms

- Angiosperms are defined by having seeds in the enclosing fruit derived from the ovary of a flower.
- The flower consists of primarily sporophytic tissues, with both male and female gametophytes which are highly reduced in size in comparison to all other land plants.
- Angiosperms also have the unique property of double fertilization, producing a usually triploid endosperm in addition to the embryo.

  - ✓ Pollen grains represent the highly reduced haploid male gametophyte generation in flowering plants, consisting of just two or three cells when released from the anthers.
  - ✓ Their role is to deliver twin sperm cells to the embryo sac to undergo fusion with the egg and central cell.
  - ✓ This double fertilization event along with the functional specialization of the male gametophyte, are considered to be key innovations in the evolutionary success of flowering plants.

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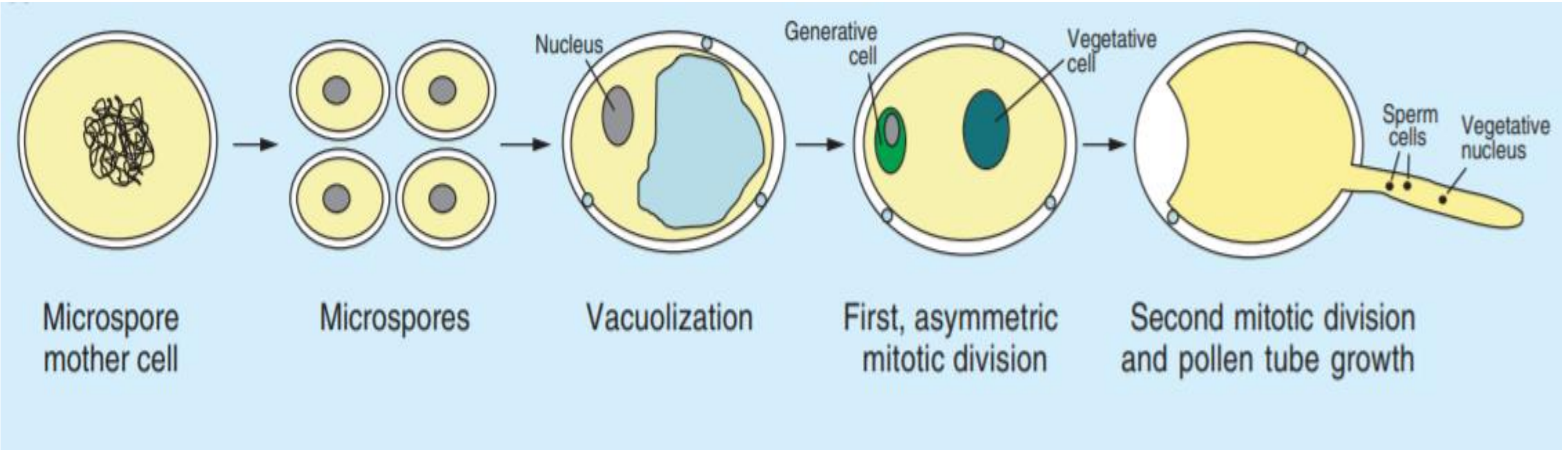
- The male gametophyte is formed in the anthers of the stamens, and the female gametophyte is located in the ovules within the pistil.
- In the anther, four pollen sacs (locules) contain numerous microspore mother cells, each of which undergoes meiosis to produce four microspores in a tetrad.
- The male gametophyte generation begins with the microspore. Initially, the microspore has a uniformly distributed cytoplasm with a centrally located haploid nucleus.



Male gametophyte. Meiosis produces four microspores; vacuolization is accompanied by nuclear migration. The microspore divides asymmetrically to form a large vegetative cell and a small generative cell. The generative cell then produces two sperm cells, which later move toward the ovule through the growing pollen tube.

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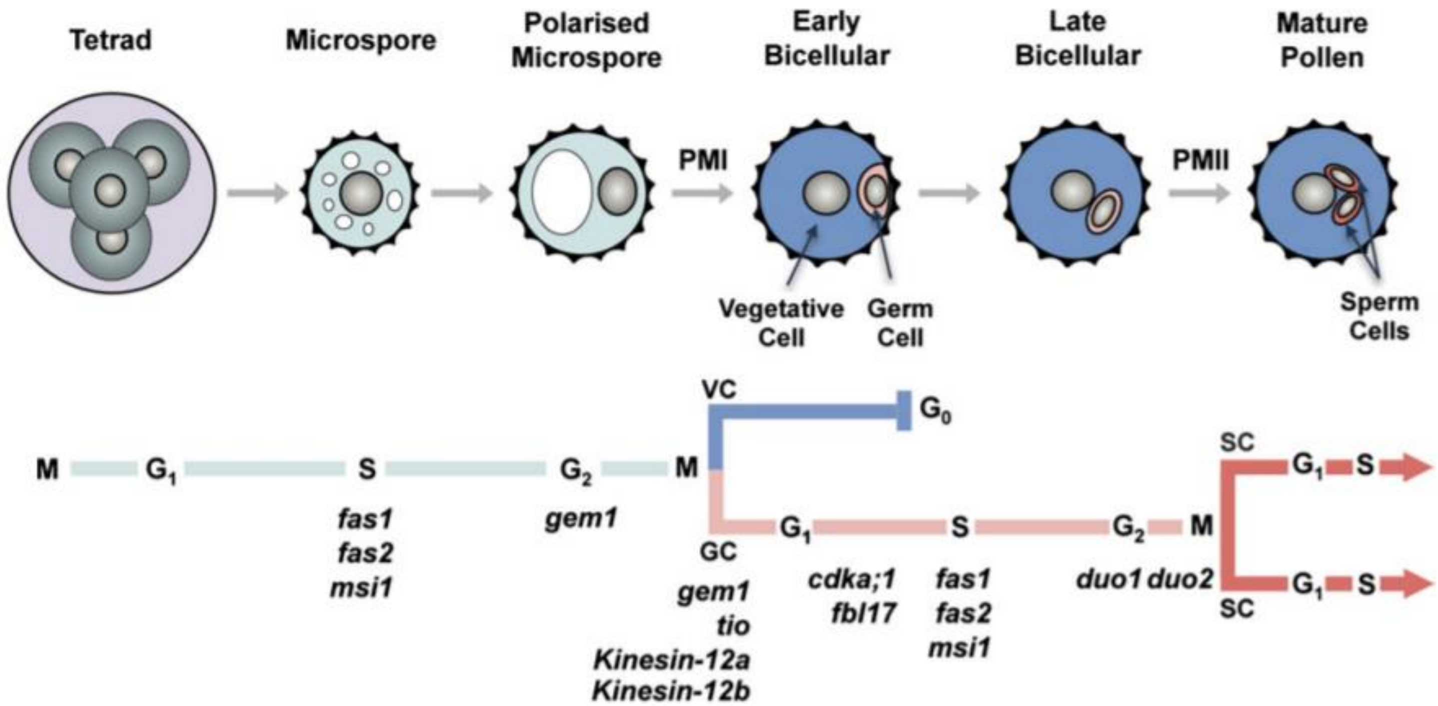

Male gametophyte development in Arabidopsis. Schematic diagram representing the distinct morphological stages of male gametophyte development in Arabidopsis along with a colour-coded timeline of the cell cycle progression of each cell type. Known mutations critical during male gametophyte development are listed below the time-line at the point that they are known to act. During microsporogenesis, microsporocytes undergo a meiotic division to produce a tetrad of four haploid microspores. During microgametogenesis, the released microspores undergo a highly asymmetric division, called Pollen Mitosis I (PMI), to produce a bicellular pollen grain with a small germ cell engulfed within the cytoplasm of a large vegetative cell. Whilst the vegetative cell exits the cell cycle, the germ cell undergoes a further mitotic division at Pollen Mitosis II (PMII) to produce twin sperm cells. The sperm cells then continue through the cell cycle to reach G2 prior to karyogamy and double fertilization. VC, vegetative cell; GC, germ cell; SC, sperm cell.

Michael Borg, Lynette Brownfield and David Twell* (2009) Male gametophyte development: a molecular perspective
Journal of Experimental Botany, Vol. 60, No. 5, pp. 1465–1478. doi:10.1093/jxb/ern355

- ✓ Induction of symmetrical division at PMI has demonstrated that vegetative cell gene expression is the default developmental pathway and that division asymmetry is critical for correct germ cell differentiation.
- ✓ After PMI, the large vegetative cell has dispersed nuclear chromatin and exits the cell cycle in G1.
- ✓ The vegetative cell nurtures the developing germ cell and gives rise to the pollen tube following successful pollination.
- ✓ This pollen tube grows through the stylar tissues of the gynoecium to deliver twin sperm cells to the embryo sac.
- ✓ During pollen maturation, the vegetative cell accumulates carbohydrate and/or lipid reserves along with transcripts and proteins that are required for rapid pollen tube growth.
- ✓ Osmoprotectants, including disaccharides, proline and glycine-betaine, which are thought to protect vital membranes and proteins from damage during dehydration, are also accumulated.
- ✓ The smaller germ cell has condensed nuclear chromatin and continues through a further round of mitosis, called Pollen Mitosis II (PMII), to produce twin sperm cells.
- ✓ In species that shed tricellular pollen, such as Arabidopsis thaliana, PMII takes place within the pollen grain prior to anthesis.
- ✓ This is in contrast to the majority of species that shed their pollen in a bicellular state, such as Lilium longiflorum, with PMII taking place in the growing pollen tube.
- ✓ Following PMII, a physical association between the sperm cells and the vegetative nucleus is established that is referred to as the male germ unit (MGU).
- ✓ Mutations affecting either the assembly (germ unit malformed or gum mutants) or positioning (MGU displaced or mud mutants) of the MGU in Arabidopsis pollen lead to reduced male transmission

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- ✓ **A large vacuole later forms at the center, displacing the nucleus to the side.**
- ✓ **In many flowering plants, including the model species Arabidopsis and maize, two mitotic divisions occur during pollen development.**
- ✓ **The first division produces a large vegetative cell and a much smaller generative cell.**
- ✓ **The vegetative cell inherits most of the cytoplasm from the microspore cell, has a relatively loose nucleus that is active in transcription, and completely envelopes the generative cells.**
- ✓ **In contrast, the generative nucleus is more tightly organized and less active in transcription.**
- ✓ **The generative cell later undergoes a second mitosis to produce two sperm cells.**
- ✓ **Pollen development depends on the function of a surrounding sporophytic tissue called tapetum.**
- ✓ **The pollen dehydrates during maturation; after pollination, the pollen grain rehydrates and germinates to produce a pollen tube.**
- ✓ **This tube grows towards the ovule, providing a passageway for the sperm cells to reach the female gametophyte.**
- ✓ **Because the female gametophyte is within the female reproductive organ of the sporophyte, the pollen tube must extend considerable distances — many times the size of the pollen grains — to reach the egg.**

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- The angiosperm gametophytes are composed of few cells and are embedded within the sexual organs of the flower.
- The female gametophyte develops within the ovule and generally consists of three antipodal cells, one central cell, two synergid cells, and one egg cell.
- The female gametophyte is also commonly called the embryo sac or megagametophyte.
- The male gametophyte, also called the pollen grain or microgametophyte, develops within the anther and consists of two sperm cells encased within a vegetative cell

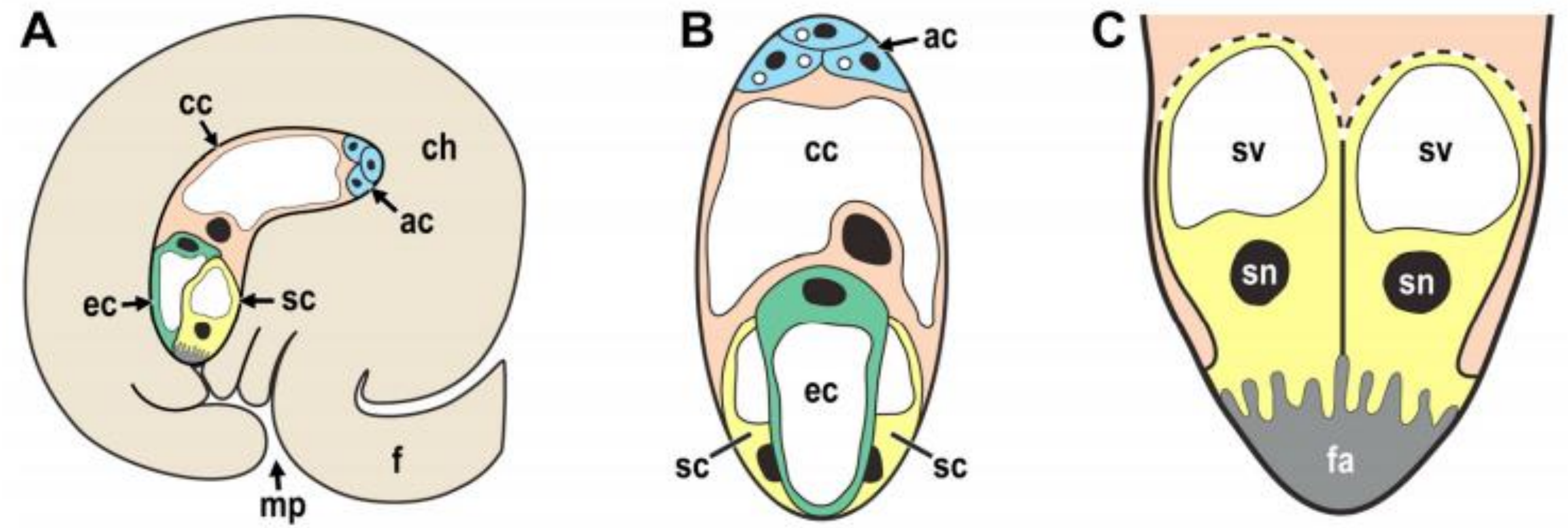


Figure 1. The Arabidopsis female gametophyte.

(A) Ovule. **(B)** Female gametophyte. **(C)** Synergid cells.

View in **(B)** and **(C)** is perpendicular to that in **(A)**. The mature female gametophyte in Arabidopsis is approximately 105 µm in length and approximately 25 µm in width. In all panels, the black circles/ovals represent nuclei and the white areas represent vacuoles. The dashed line at the chalazal ends of the synergid cells in **(C)** represents a discontinuous or absent cell wall. Abbreviations: ac, antipodal cells; cc, central cell; ch, chalazal region of the ovule; ec, egg cell; f, funiculus; fa, filiform apparatus; mp, micropyle; sc, synergid cell; sn, synergid nucleus, sv, synergid vacuole.

- **Female gametophyte formation is required for sexual and asexual seed development in angiosperms.**

- **In sexually reproducing angiosperms, seed formation begins when pollen is transferred from the anther to the carpel's stigma.**

- **The male gametophyte then forms a pollen tube that grows through the carpel's internal tissues and into the ovule to deliver its two sperm cells to the female gametophyte.**

- **One sperm fertilizes the egg, and the second fuses with the central cell.**

- **Following double fertilization, the egg cell gives rise to the seed's embryo, which is the beginning of the sporophyte generation, the central cell develops into the seed's endosperm, which surrounds and provides nutrients to the developing embryo, and the surrounding sporophytic cells give rise to the seed coat.**

- **Plants can also produce seeds asexually by apomixis.**

- **Apomixis occurs in over 40 plant families and more than 400 genera.**

- **Apomixis does not occur in Arabidopsis but is found in a related genus, Boechera.**

- **Apomictic species exhibit much variation in the developmental mechanism leading to asexual seed production, and some routes bypass female gametophyte formation.**

- **A form of apomixis that involves the female gametophyte is gametophytic apomixis During gametophytic apomixis, meiotic reduction is bypassed and diploid female gametophytes are formed by a variety of developmental routes in different species.**

- **The egg cell then forms an embryo autonomously (i.e., without fertilization; also termed parthenogenesis) and endosperm formation may occur autonomously or may require central cell fertilization (termed pseudogamy).**

- **Apomixis gives rise to clonal progeny with a maternal genotype through seed. Apomixis, therefore, could be important in plant breeding to fix hybrid vigor and could significantly reduce the costs of generating high yielding hybrid seeds.**

- **Analysis of female gametophyte development is therefore important for many reasons.**

- **It is integral to the plant life cycle and essential for both sexual and apomictic seed formation.**

- **The female gametophyte controls many steps of the angiosperm sexual reproductive process: during pollen tube growth and fertilization, the female gametophyte guides the pollen tube to the ovule and embryo sac, controls pollen tube growth within the female gametophyte, and mediates fertilization of the egg cell and central cell; and upon double fertilization, female gametophyte-expressed genes participate in inducing embryo and endosperm formation during seed development.**

- **In the absence of fertilization in autonomous apomicts, the unreduced female gametophyte contains factors to stimulate embryo and endosperm formation.**

FEMALE GAMETOPHYTE DEVELOPMENT IN ARABIDOPSIS

- Angiosperms exhibit many different patterns of female gametophyte development.
- Arabidopsis undergoes the Polygonum-type pattern, which is the most common pattern and is exhibited by over 70% of flowering plants.
- The Polygonum-type pattern is also exhibited by many economically important groups including Gramineae (e.g., maize, rice, wheat), Phaseoleae (e.g., beans, soybean), Brassicaceae (e.g., Brassica), Malvaceae (e.g., cotton), and Solanaceae (e.g., pepper, tobacco, tomato, potato, petunia), as well as most apomictic species.
- Regardless of the specific developmental pattern, female gametophyte development occurs within the developing ovule and consists of two main phases: megasporogenesis followed by megagametogenesis.
- During Arabidopsis megasporogenesis, the diploid megaspore mother cell undergoes meiosis and gives rise to haploid megaspores.
- During Arabidopsis megagametogenesis, one of the megaspores develops into the mature female gametophyte

Polygonum-type pattern : A single nucleus undergoes two rounds of mitosis, producing a four-nucleate cell with two nuclei at each pole. ... The monosporic, Polygonum type of female gametophyte is typically a seven-celled structure at maturity.

- **In Arabidopsis and most other species, the archesporial cell develops directly into the megaspore mother cell.**

- **In some flowering plants, the archesporial cell enlarges and undergoes a periclinal division, and subsequently the inner cell differentiates into the megaspore mother cell.**

- **In Arabidopsis and most other species, one meiotic product contributes to formation of the mature female gametophyte and this pattern is referred to as monosporic.**

- **Angiosperms exhibit two other megasporogenesis patterns referred to as bisporic and tetrasporic.**

- **In the bisporic pattern, cell plates form following meiosis I but not meiosis II and one of the two-nucleate megaspores degenerates, resulting in a single functional megaspore.**

- **In the tetrasporic pattern, cell plates fail to form following both meiotic divisions, resulting in one four-nucleate megaspore.**

- **Angiosperm species also exhibit significant variation in megagametogenesis.**

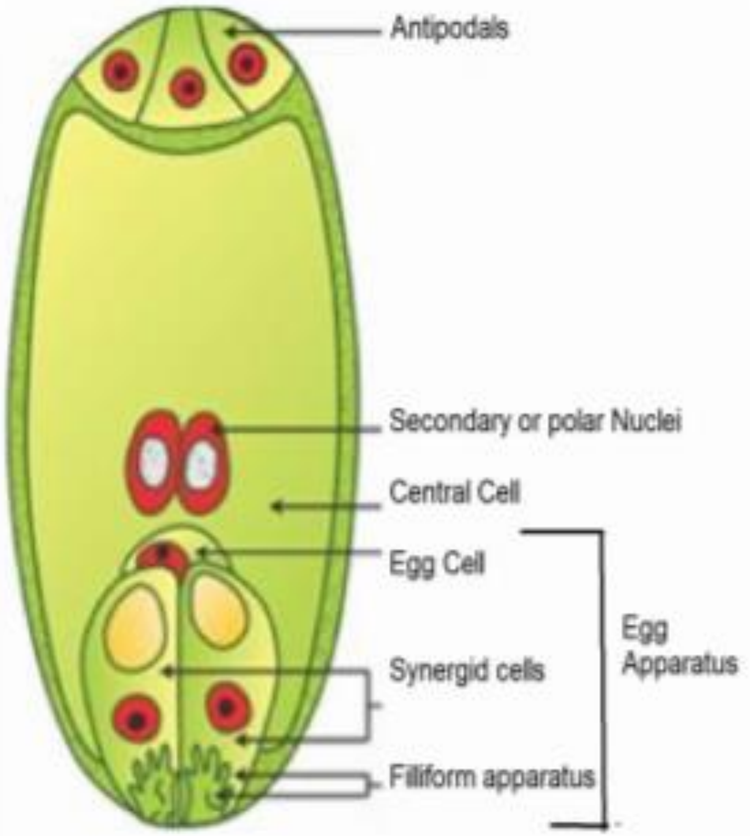
- **Most species undergo the same general pattern as for Arabidopsis: a phase of nuclear proliferation without cytokinesis followed by cellularization and differentiation.**

- **Variation arises due to the number of nuclei within the megaspore that gives rise to the female gametophyte (i.e., the type of megasporogenesis), the number of mitoses prior to cellularization, the timing of fusion of the polar nuclei, and whether or not additional mitoses occur after cellularization.**

- **For example, in maize, which has a Polygonum-type female gametophyte, the polar nuclei do not fuse until fertilization and the antipodal cells proliferate into 40 or more cells.**

EMBRYO SAC (FEMALE GAMETOPHYTE)

- Functional megaspore represents the first cell of the female gametophyte.
- Mostly chalazal and grows along the micropylar- chalazal axis.
- The nucleus undergoes three mitotic divisions and form eight nuclei.
- These nuclei are reorganised; one nucleus from each group at a pole migrates to the centre of the cell, called polar nuclei.
- The three nuclei left at the chalazal end are surrounded by walls and form antipodals.
- The three nuclei located at the micropylar endconstitute the egg apparatus, one serve as egg or female gamete and the other two as synergids.
- The whole structure with two polar nuclei, three antipodals, one egg and two synergids is the mature female gametophyte or embryo sac.



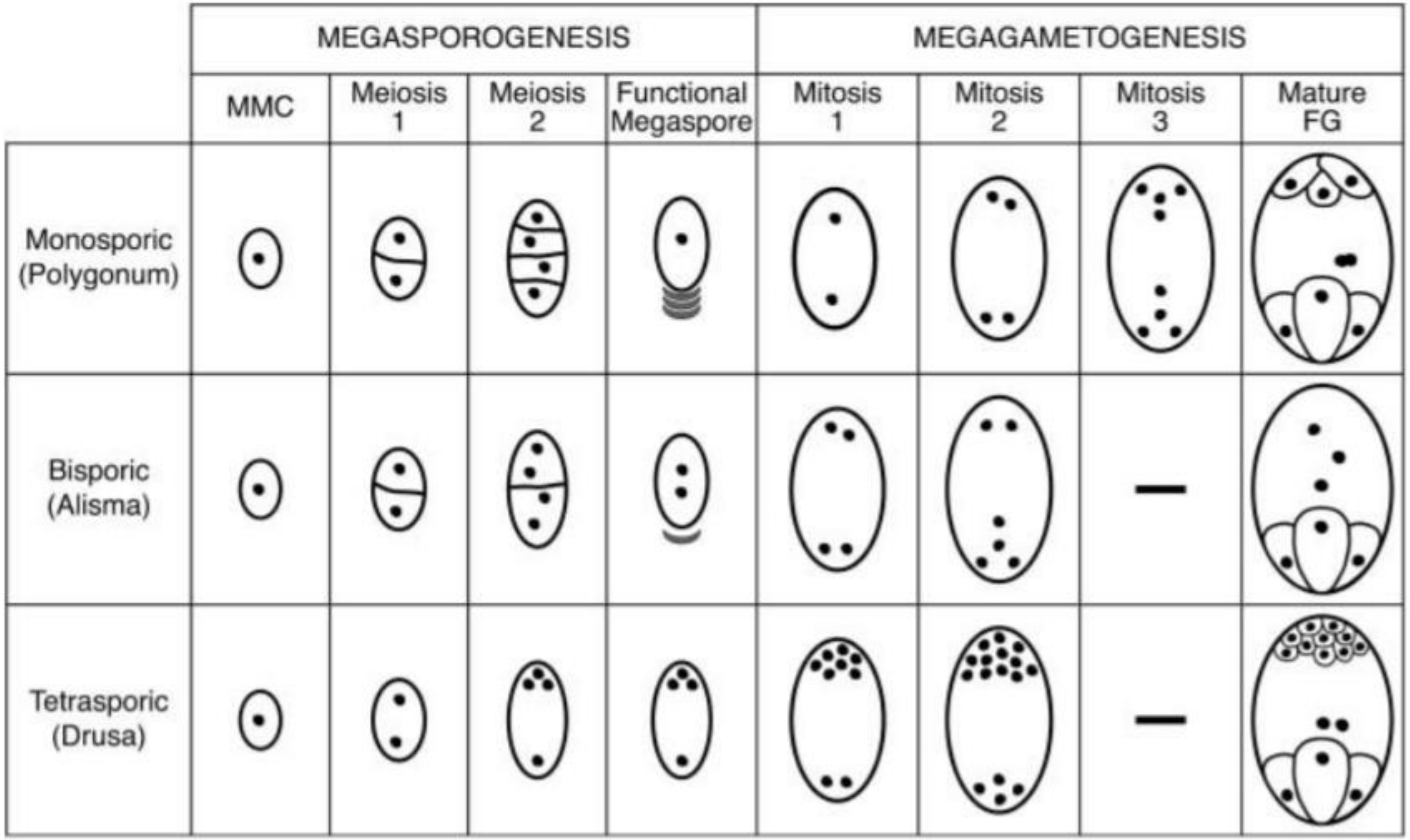


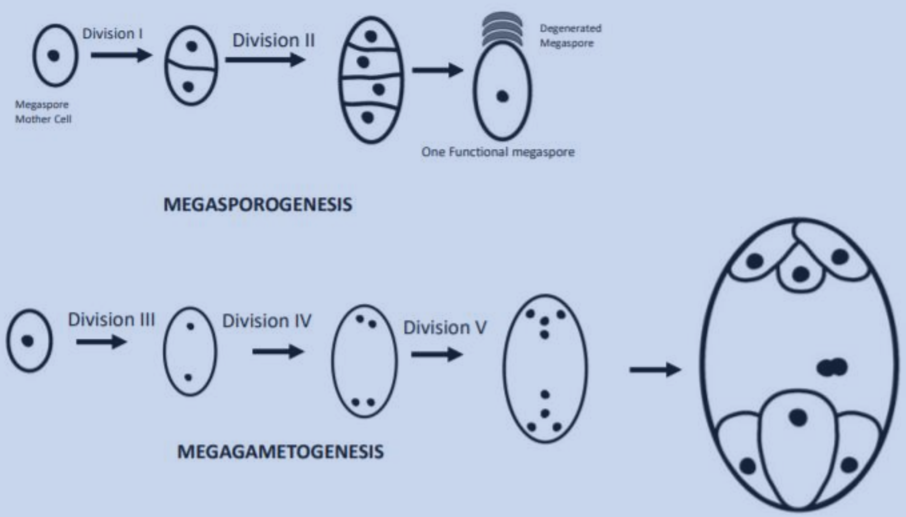
Figure Patterns of Female Gametophyte Development Exhibited by Angiosperms. Genera exhibiting these patterns are indicated in parentheses. More comprehensive descriptions of the variation among angiosperms can be found in several reviews. In this figure, the chalazal end of the female gametophyte is up and the micropylar end is down. FG, female gametophyte.

TYPE OF EMBRYO SAC (FEMALE GAMETOPHYTE)

1. Monosporic embryo sac
2. Bisporic embryo sac
3. Tetrasporic embryo sac

- **Basis for classification**
  - The number of megaspores taking part in the development of embryo sac
  - The number of divisions occurring in the nucleus of the functional megaspore
  - Organization of nuclei in the mature embryo sac

Polygonum type of Embryo Sac



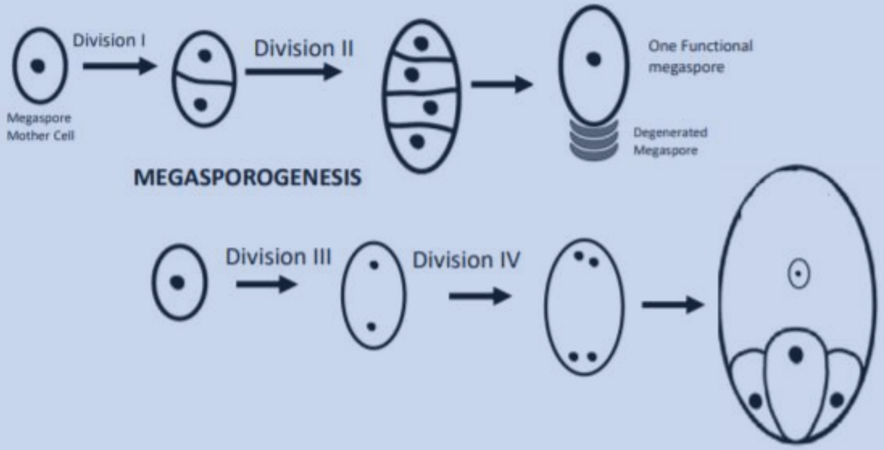
MONOSPORIC EMBRYO SAC

Monosporic embryo sac develops from a single megaspore and as such all the nuclei present in this type of embryo sac are genetically similar.

Types of Monosporic Embryo Sac

- **Polygonum type**
  - 8 nucleated
  - Most common type (81% families)
  - First time described in *Polygonum divaricatum* by Strasburger (1879)
  - Develops from the chalazal megaspore.
  - The nucelus is divides thrice to form eight nuclei embryo sac

- **Oenothera type:**
  - 4 nucleated embryo sac (Egg apparatus- 3 cells and Polar nucleus- single cell)
  - Develops from micropylar megaspore
  - Does not have antipodals
  - Example – Onagraceae family

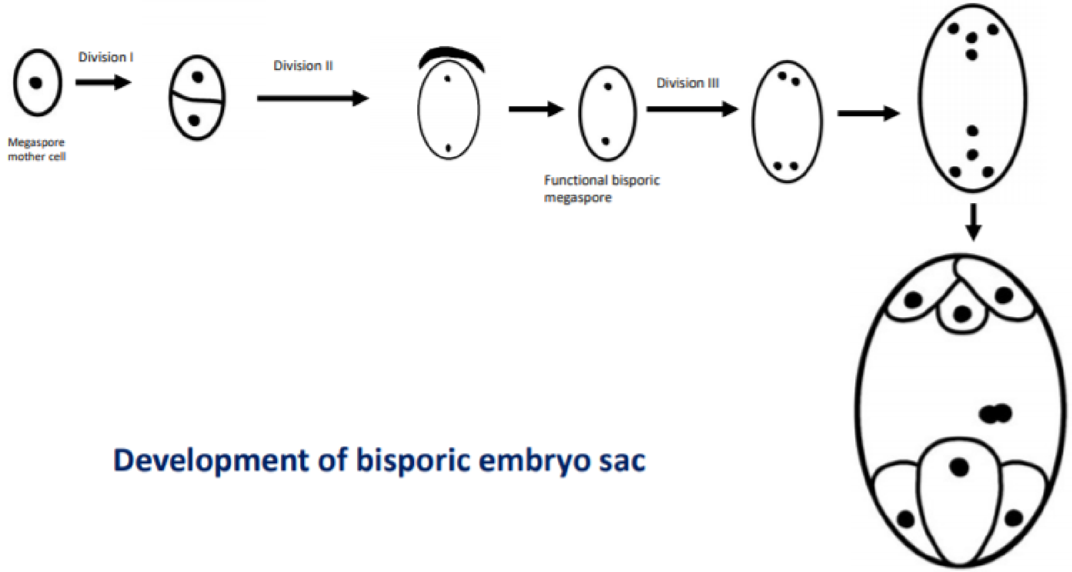


BISPORIC EMBRYO SAC

- Develops from one of the two dyads formed as a result of the first meiotic division (Meiosis I) of Megaspore Mother Cell.
- Both the nuclei of the functional dyad take part in the formation of embryo sac.
- Each nucleus undergoes two mitotic divisions and the mature embryo sac is 8 nucleated.
- The eight nuclei are organised into antipodals, egg apparatus and polar nuclei as in *Polygonum* type of embryo sac.
- The 4 nuclei derived from one megaspore nucleus are genetically different from the other four derived from the second megaspore nucleus.

On the basis of the position of functional dyad bisporic embryo sacs are of two types

ALLIUM TYPE: Develops from the chalazal dyad
ENDYMION TYPE: Develops from the micropylar dyad.



Development of bisporic embryo sac

TETRASPORIC EMBRYO SAC

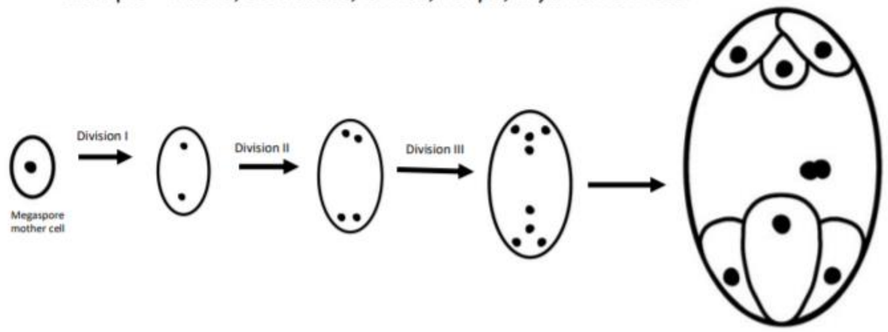
- Meiotic division of the megaspore mother cell is not accompanied by cytokinesis and hence all the four haploid nuclei lie in a single cell called **Coeno-megaspore**.
- All four nuclei of coeno-megaspore participate in the formation of embryo sac
- Genetically more heterogeneous than bisporic type of embryo sac.
- The tetrasporic embryo sacs are further divided on the basis of following criteria-
  - The position of haploid nuclei in the coeno-megaspore
  - The number of times these nuclei divide
  - Organization of nuclei in the mature embryo sac

TYPE OF TETRASPORIC EMBRYO SAC

- No nuclear fusion occurs
  - Adoxa Type
  - Plumbago Type
  - Penaea Type
  - Pepromia Type
  - Drusa Type
- After the second meiotic division three megaspore nuclei fuse to form triploid nucleus at the chalazal end of the coenomegaspore, The fourth nucleus at micropylar end remains haploid
  - Fritillaria Type
  - Plumbagella Type

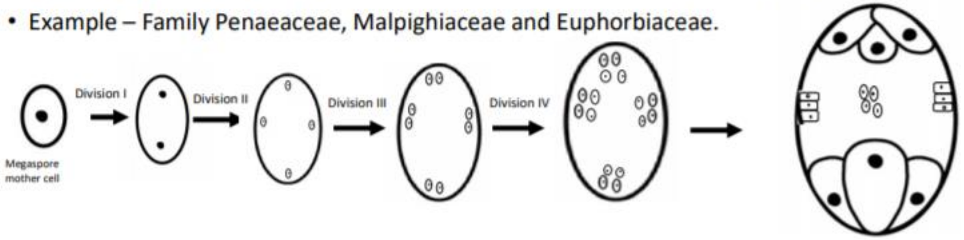
ADOXA TYPE

- It has 8 nuclei which are formed by the mitotic division of the four haploid nuclei of the coeno-megaspore.
- The arrangement of the 8 nuclei in the embryo sac is the same as in Polygonum type.
- Example – *Adoxa, Sambucus, Ulmus, Tulipa, Erythronium* etc.



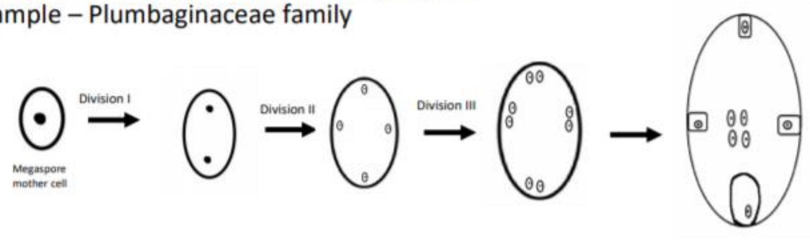
PENAEA TYPE

- The four haploid nuclei of the coeno-megaspore undergo two successive mitotic divisions forming 16 nuclei.
- These nuclei arrange themselves in four groups of four each, one at the micropylar end, one at chalazal end and one each on the two lateral sides.
- Now one nucleus from each groups migrates to the centre, and these four nuclei in the centre form polar nuclei.
- The three nuclei at the micropylar end are cut off by membranes and form the egg apparatus.
- The remaining three groups of nuclei (one chalazal and two lateral) degenerate at maturity.
- Highly polyploid (5x) primary endosperm nucleus is formed after double fertilization.
- Example – Family Penaeaceae, Malpighiaceae and Euphorbiaceae.



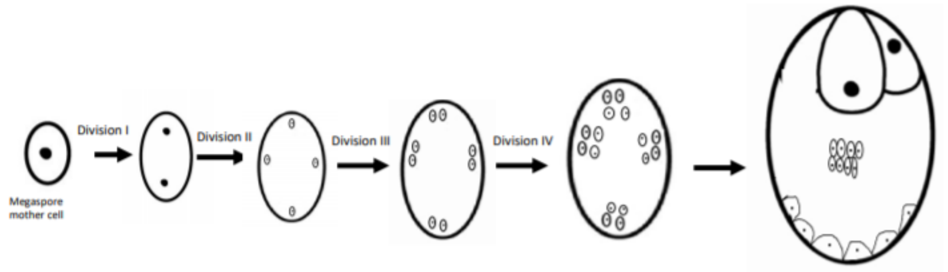
PLUMBAGO TYPE

- This type of embryo sac is characterized by the absence of synergids and antipodals.
- Out of four haploid coeno-megaspore one migrates to the micropylar end, one at chalazal end and two at the lateral sides.
- Each nuclei divides again and formed four groups of two nuclei.
- One of the nucleus from each group moves to the center of the cell and form four polar nuclei.
- The remaining nucleus at the micropylar is cut off by a membrane and form the egg. There are no synergids.
- The other three nuclei usually disappear but occasionally they too may be cut off by membranes and appear as accessory egg cells.
- Example – Plumbaginaceae family



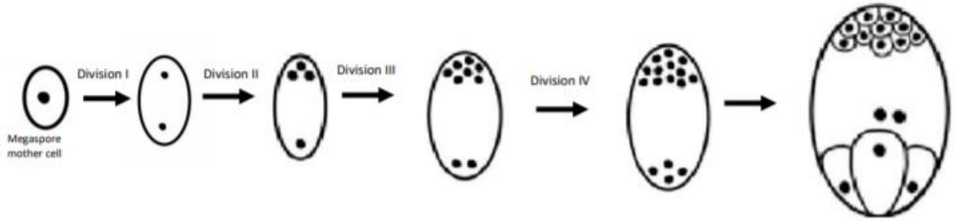
PEPEROMIA TYPE

- The egg apparatus of Peperomia type is characterized by a single synergid.
- The four haploid nuclei of coeno-megaspore undergo two successive mitotic divisions forming 16 nuclei.
- Two nuclei at the micropylar end form egg and a synergid, eight fuse in the centre of the cell to form a polar nucleus and the remaining six at the chalazal end formed antipodals.
- Example- Peperomia and Gunnera



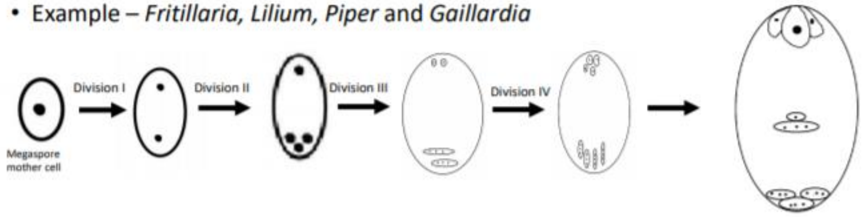
DRUSA TYPE

- 16 nucleate embryo sac
- This type of embryo sac is characterised by large number of antipodals
- In the mature embryo sac three nuclei form egg apparatus. Two act as polar nuclei and the remaining 11 nuclei are cut off by membrane and form antipodal cells.
- The number and organization of nuclei may vary due to irregularity in the divisions.
- Example – *Drusa, Rubia, Chrysanthemum, Ulmus* etc.



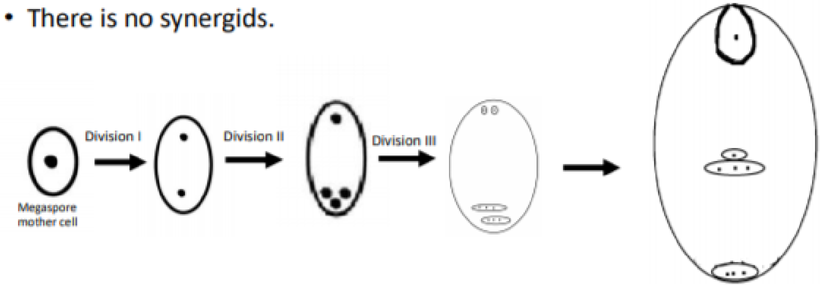
FRITILLARIA TYPE

- The four haploid nuclei of the coeno-megaspore arrange themselves in two groups – three at the chalazal end in the form of a triploid nucleus and one haploid at the micropylar end.
- The triploid chalazal as well as the haploid micropylar nucleus undergo two mitotic divisions and as a result four trploid nuclei are formed at the chalazal end and four haploid at the micropylar end.
- In mature embryo sac three haploid nuclei organize into egg apparatus, three triploid into antipodal and remaining one haploid and one triploid nuclei move to the centre where they fuse to form a tetraploid polar nucleus.
- Example – *Fritillaria, Lilium, Piper* and *Gaillardia*



PLUMBAGELLA TYPE

- The initial development is similar to Fritillaria type and a triploid nucleus is formed at the chalazal end and a haploid at the micropylar end.
- Each of these nuclei undergoes a single mitotic division and form two groups of two nuclei each.
- One triploid nuclei from chalazal end and one haploid nucleus from the micropylar end fuse at the centre and form tetraploid polar nucleus.
- One haploid nucleus at the micropylar end forms the egg and one triploid nucleus at the chalazal end the single antipodal.
- There is no synergids.



- Megasporogenesis comprises three major events:
- megaspore mother cell formation,
- meiosis to produce haploid megaspores, and
- megaspore selection (i.e., selection of the megaspore that develops into the female gametophyte).

- Ovule primordia in Arabidopsis arise as finger-like projections from the placental tissue of the ovary.
- During early ovule development, a sub-epidermal cell at the distal end of the ovule primordium forms the archesporial cell.
- In Arabidopsis and most other species, the archesporial cell differentiates directly into the megaspore mother cell (also called the female meiocyte or megasporocyte); thus, in these species, there is no functional difference between an archesporial cell and a megaspore mother cell.
- The ovule cells that do not develop into the megaspore mother cell are sporophytic cells and are often referred to as somatic cells.
- Relative to the somatic cells, the megaspore mother cell is larger and has a denser cytoplasm and a larger nucleus.
- Just before meiosis, the megaspore mother cell is dramatically enlarged and elongated.
- The megaspore mother cell then undergoes meiosis and gives rise to four one-nucleate, haploid megaspores.
- Subsequently, three of the megaspores degenerate and one survives.

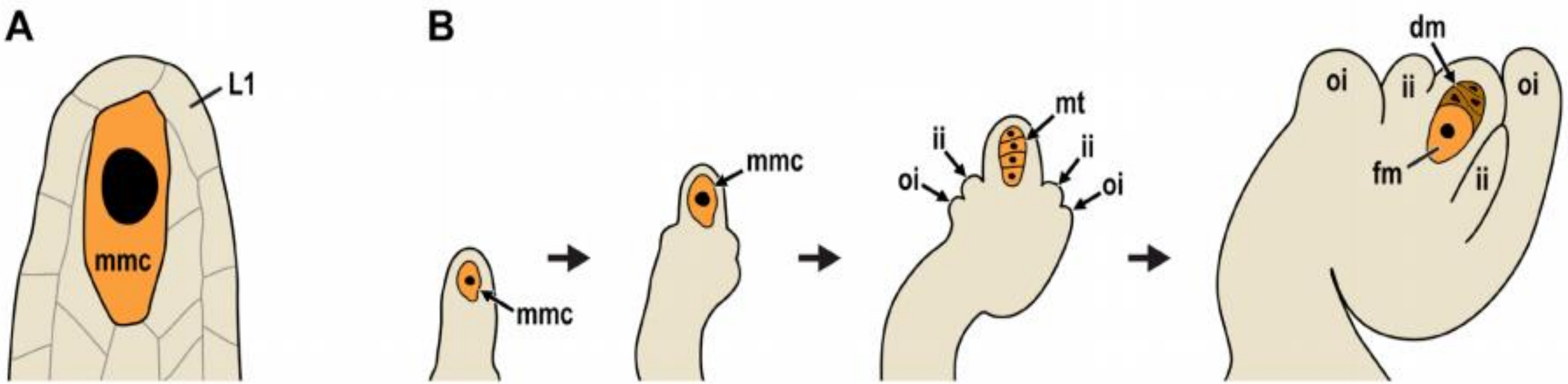


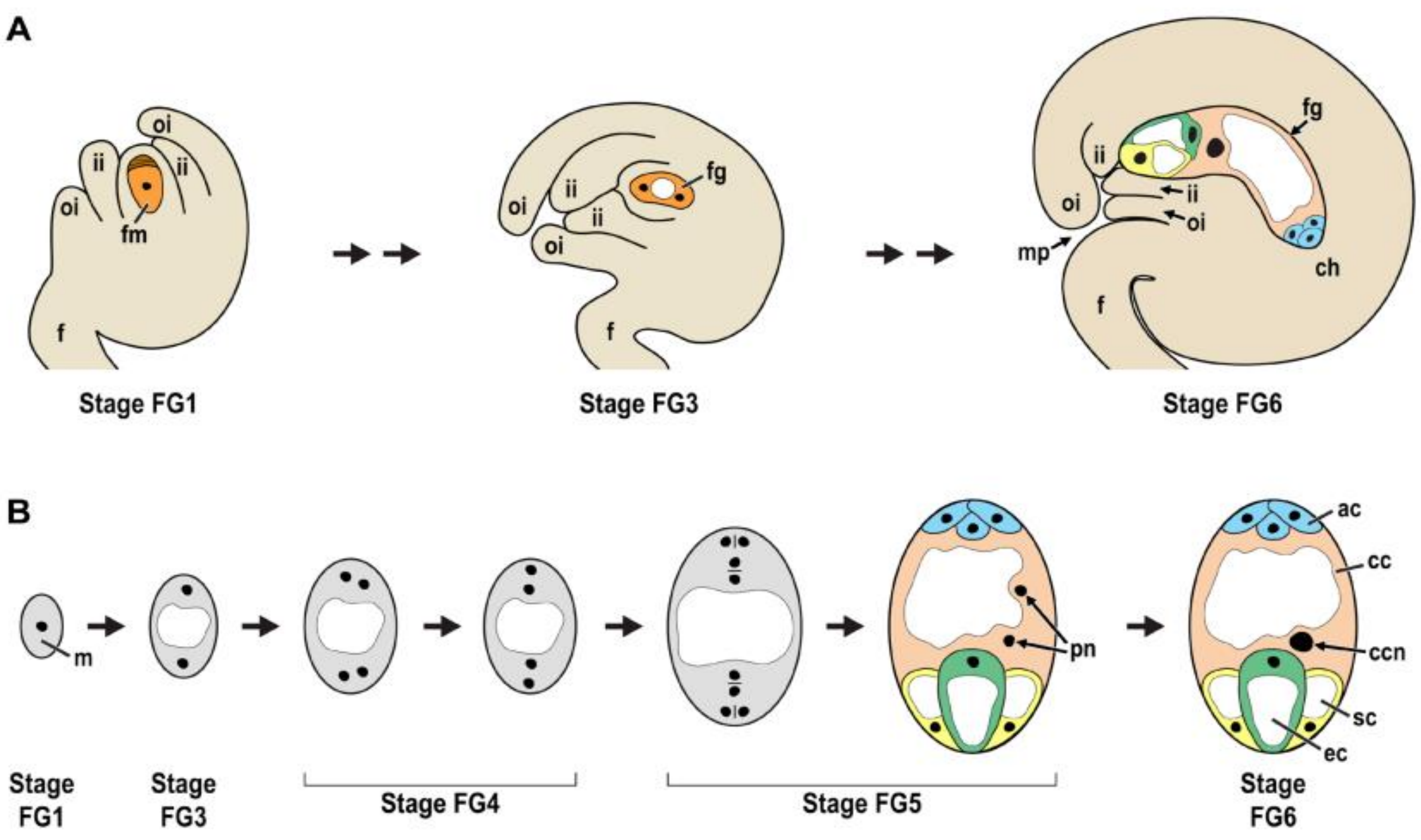
Figure 2. Megasporogenesis in Arabidopsis.

(A) Apical region of a young, finger-like ovule primordium. The megaspore mother cell forms from a sub-epidermal cell at the distal end of the ovule primordium. L1 is the outer layer of cells.

(B) Steps of megasporogenesis. Ovule primordia arise as finger-like projections from the placenta. The megaspore mother cell undergoes meiosis and forms four megaspores. Three of the megaspores undergo cell death. The chalazal-most megaspore survives, becomes the functional megaspore, and undergoes megagametogenesis.

Black circles/ovals represent nuclei.

Abbreviations: dm, degenerating megaspores; fm, functional megaspore; ii, inner integument; L1, L1 epidermal layer of the ovule primordium; mmc, megaspore mother cell; mt, meiotic tetrad; oi, outer integument.



Megagametogenesis in Arabidopsis.

(A) Steps of megagametogenesis emphasizing development within the ovule.

(B) Stages of megagametogenesis The megaspore contains a single nucleus (stage FG1). This nucleus undergoes two rounds of mitosis, producing a four-nucleate coenocyte, with two nuclei at each pole separated by a large central vacuole (stage FG4). During a third mitosis, phragmoplasts and cell plates form between sister and non-sister nuclei and the nuclei become completely surrounded by cell walls (Stage FG5).

During cellularization, the polar nuclei migrate toward the center of the female gametophyte and fuse before fertilization. These events produce a seven-celled structure consisting of three antipodal cells, one central cell, two synergid cells, and one egg cell. If the female gametophyte is not fertilized, the antipodal cells eventually degenerate (Stage FG7, not shown). White areas represent vacuoles and black circles/ovals represent nuclei. Abbreviations: ac, antipodal cells; cc, central cell; ccn; central cell nucleus; ch, chalazal region of the ovule; ec, egg cell; f, funiculus; fg, female gametophyte; fm, functional megaspore; ii, inner integument; m, megaspore; mp, micropyle; oi, outer integument; pn, polar nuclei; sc, synergid cells.

- **In Arabidopsis and most other species, the chalazal-most megaspore survives during megaspore selection.**

- **Programmed cell death is likely to be the cause of megaspore degeneration because TUNEL assays show DNA fragmentation in degenerating megaspores of alfalfa (Medicago sativa L) ovules.**
- **Furthermore, degenerating megaspores express MPS-ONE-BINDER (MOB1) genes, which encode homologs of fly and mammalian proteins that regulate apoptosis factors.**
- **A histological marker associated with the megaspore mother cell is callose (β-1,3-glucan).**
- **During megasporogenesis, the megaspore mother cell and the cells undergoing meiosis accumulate callose in their cell walls.**
- **After meiosis, callose (function not understood) is lost in the cell walls of the selected megaspore as it enlarges and begins the transition to megagametogenesis.**

- **Megagametogenesis:**

- **Megagametogenesis also involves three identifiable events: a series of mitoses without cytokinesis, followed by cellularization of the nuclei and then cell differentiation.**

- **In Arabidopsis, the single surviving megaspore enlarges and then undergoes two rounds of mitosis without cytokinesis, resulting in a four-nucleate coenocyte with two nuclei at each pole.**

- **During a third mitosis, phragmoplasts and cell plates form between sister and non-sister nuclei; this is the beginning of the cellularization process and the female gametophyte cells quickly become completely surrounded by cell walls.**

- **During and after cellularization, one nucleus from each pole (the polar nuclei) migrates toward the center of the developing female gametophyte and they fuse.**

- **These events result in a seven-celled structure consisting of three antipodal cells, one central cell, two synergid cells, and one egg cell.**

- **The central cell inherits two identical haploid nuclei and is therefore homodiploid.**

- **The other cells all inherit single haploid nuclei. If the female gametophyte is unfertilized, the antipodal cells eventually disappear or undergo cell death; however, at the time of fertilization, the female gametophyte most likely is a seven-celled structure (i.e., the antipodal cells are present).**

STRUCTURE OF THE MATURE FEMALE GAMETOPHYTE

- **The egg and central cells are polarized such that the nuclei of both cells lie very close to each other.**

- **This feature is important for double fertilization because these two nuclei are the targets of the two sperm nuclei.**

- **Furthermore, in the regions where the egg, synergid, and central cells meet, the cell walls are absent or discontinuous and the plasma membranes of these cells are in direct contact with each other.**

- **The absent cell walls in this region provide direct access of the sperm cells to the fertilization targets because the pollen tube releases its two sperm cells into one of the synergid cells.**

- **The synergid cell wall is further specialized.**

- **At the micropylar pole, the synergid cell wall is thickened and extensively invaginated, forming a structure referred to as the filiform apparatus.**

- **The filiform apparatus greatly increases the surface area of the plasma membrane in this region and contains a high concentration of secretory organelles, suggesting that it may facilitate transport of substances into and out of the synergid cells.**

- **Based on cytological staining properties in species other than Arabidopsis, the filiform apparatus appears to be composed of a number of substances including cellulose, hemicellulose, pectin, callose, and protein.**

- The filiform apparatus has at least two functions associated with the fertilization process.

- First, the synergid cells secrete pollen tube attractants via the filiform apparatus.

- In addition, the pollen tube enters the synergid cell by growing through the filiform apparatus, suggesting that the filiform apparatus is important for pollen tube reception.

- By contrast, the antipodal cells in Arabidopsis have no dramatic specializations and no known function.

- In other species, the antipodal cells contain finger-like cell wall projections resembling the filiform apparatus.

- In cereals, the antipodal cells proliferate into as many as 100 cells.

- These observations suggest that the antipodal cells indeed have a function and that they may function as transfer cells, transporting substances from the surrounding ovule cells into the female gametophyte.

- Female Gametophyte Polarity:
- The ovule and female gametophyte are polarized structures.
- The ovule's micropylar pole is the end at which the integuments form a pore, and its chalazal pole is the end that joins the funiculus.
- Within the female gametophyte, the egg and synergid cells occupy the micropylar pole and the antipodal cells lie at the chalazal pole.
- This polarity is important for fertilization because the pollen tube reaches the female gametophyte by growing through the micropyle.
- This polarity is apparent throughout female gametophyte development.
- The megaspore mother cell's cytoplasm is polarized along the chalazal-micropylar axis.
- During megaspore selection, the chalazal most megaspore survives, whereas the other three undergo cell death.

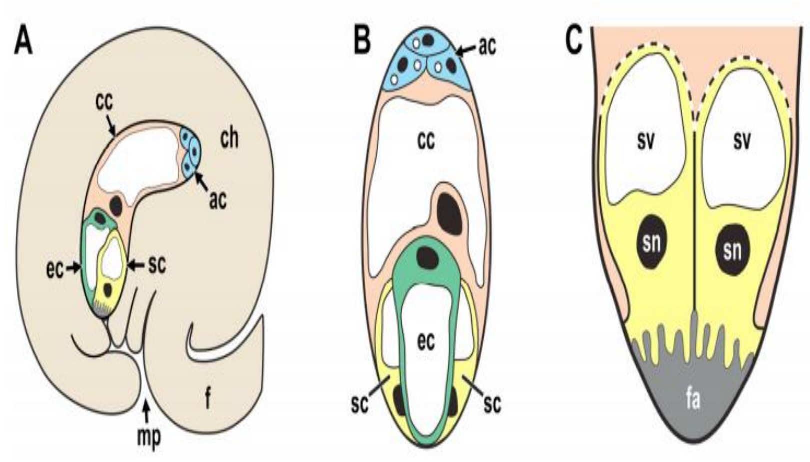


Figure 1. The Arabidopsis female gametophyte.

(A) Ovule. **(B)** Female gametophyte. **(C)** Synergid cells.

View in **(B)** and **(C)** is perpendicular to that in **(A)**. The mature female gametophyte in Arabidopsis is approximately 105 µm in length and approximately 25 µm in width. In all panels, the black circles/ovals represent nuclei and the white areas represent vacuoles. The dashed line at the chalazal ends of the synergid cells in **(C)** represents a discontinuous or absent cell wall. Abbreviations: ac, antipodal cells; cc, central cell; ch, chalazal region of the ovule; ec, egg cell; f, funiculus; fa, filiform apparatus; mp, micropyle; sc, synergid cell; sn, synergid nucleus, sv, synergid vacuole.

- During cell differentiation, the three cells at the micropylar end develop into the egg and the two synergid cells, whereas those at the chalazal end develop into three antipodal cells.
- The individual cells are polarized at the sub-cellular level.
- Female gametophyte polarity corresponds to the overall polarity of the ovule, suggesting that this polarity is regulated by the surrounding sporophytic tissue.
- Auxin gradients established by the surrounding sporophytic tissue are critical for establishing the asymmetric structure of the female gametophyte in Arabidopsis

Ovule

- **The ovule is part of the makeup of the female reproductive organ in seed plants.**

- **It's the place where female reproductive cells are made and contained, and it is what eventually develops into a seed after *fertilization*, only for the seed to then ripen and produce a complete adult plant.**

- **Ovules are contained in ovaries at the bottom of a vase-like structure, the carpel, which has a neck called a style and an opening at the top, called a stigma.**

- **After fertilization the ovule starts to swell and its wall starts to toughen up in preparation to become a seed, while the ovary starts to grow around it and becomes the fruit.**

- **Keep in mind that some plants, like the avocado, have a single ovule in their ovary, while others, like the kiwifruit, have many, which develop into many seeds in the fruit.**

- **Another way that plants differ with regards to their ovules is the place where the ovules are found.**

- **Specifically, in *gymnosperms*, such as conifers, the ovules are found on the scales of female cones, while in *angiosperms*, which are flowering plants, the ovules are found inside of the ovary within the carpel.**

➢ Components of Ovules

The ovule is made up of the nucellus, the integuments that form the outermost layer, and the female gametophyte (called an embryo sac in flowering plants), which are found at the very center.

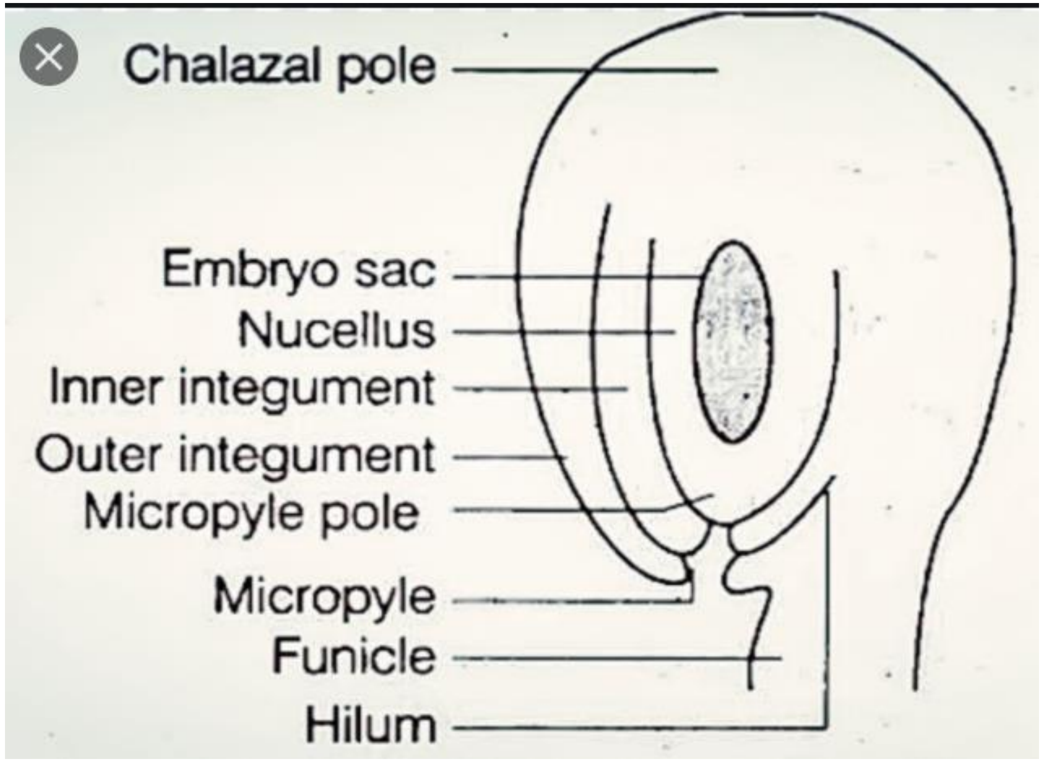
- ➢ Each ovule is attached to ovary through funicle or funiculus
- ➢ Part attaching ovule to funicle is called hilium.

➢ The Nucellus

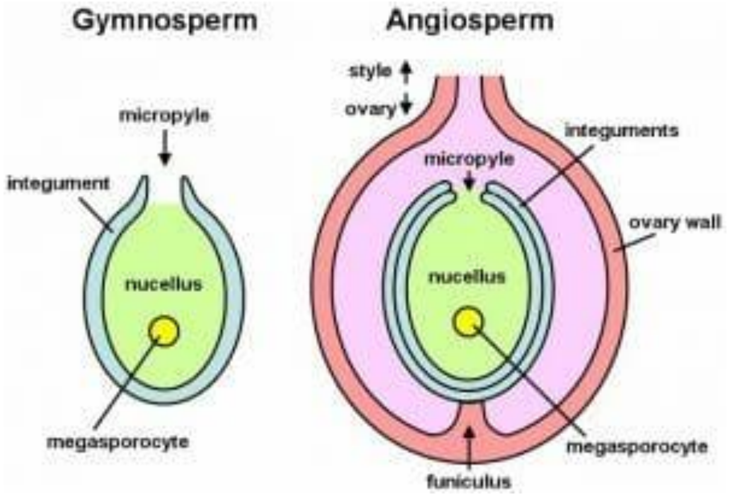
Major cells are parenchymatous called nucellus, it is the largest part of the ovule. It houses the embryo sac as well as nutritive tissue and actually remains present in some flowering plants after fertilization as a source of nutrients for the embyo. It is enclosed by layers called integuments.

➢ The Integuments

The integument is the tough outer protective layer of the ovule. Outer is called outer integument while inner is called inner integument.



- ❖ In Lorenthus, sandal etc. ovules are without integument and are known as Untegmic/Ategmic.
- ❖ In the diagram, in gymnosperms, such as pine trees and spruce trees, usually have one integument in an ovule, so we call them unitegmic.
- ❖ In sunflower, marigold etc. also only one integument is present so they are also unitegmic.
- ❖ Angiosperms, like maples and daisies, typically have two integuments, and we call them bitegmic. The integument encloses the nucellus except for a small gap, which is called the *micropyle*.



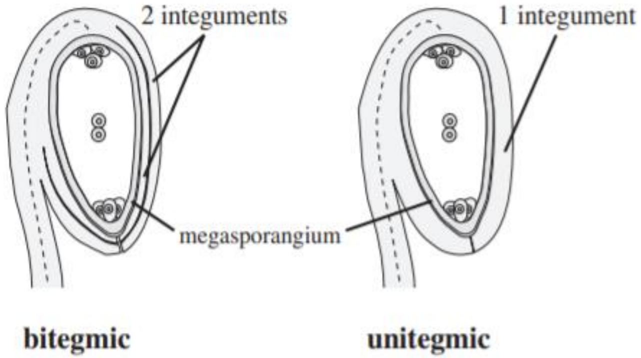


FIGURE 11.12 Ovule integument types.

- ➢ At the apex part of ovule, this covering is incomplete, leading to formation of small pore called micropyle.
- ➢ The base of ovule from where these integuments arise is called chalaza.
- ➢ In the center, a sac like structure is called embryo sac. Collectively with single egg, two synergids, 1 polar nuclei (2n) and three antipodals this structure is known as egg apparatus.

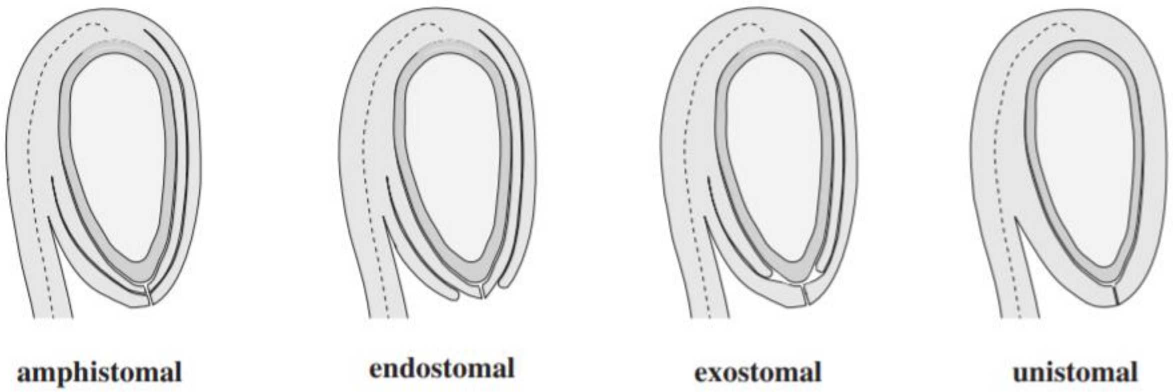


FIGURE 11.13 Ovule micropyle types.

The Female Gametophyte

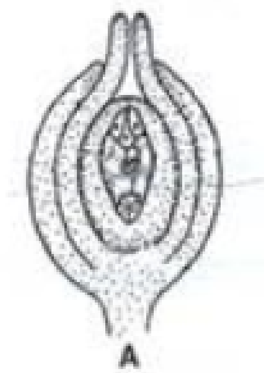
This is the part of the ovules that contains the gamete-producing sex organs, which are critical for sexual reproduction. The female gametophyte contains a single set of unpaired chromosomes, meaning that it's haploid. It is commonly called the embryo sac or *megagametophyte*.

Types of Ovules

On the basis of micropyle, chalaza and funicle, ovules can be categorized into six types

A- Orthotropous (Atropous)

- **The body of these ovules is straight**
- **The chalaza, where the nucellus and integuments merge, the *funicle*, which attaches the ovule to the placenta, and the *micropyle* are all aligned in straight line.**
- **Eg., Polygonum, Rumex**

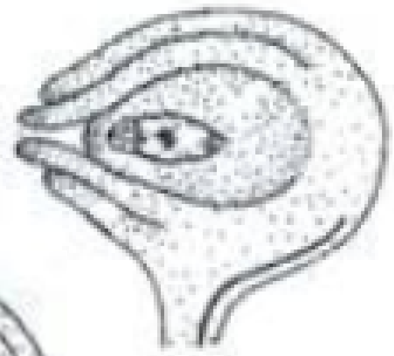


B- Anatropous

- **The ovules become completely inverted during development due to excessive growth of funicle such thay micropyle comes close to funicle and hilium.**
- **In these micropyle and chalaza are in straight line.**
- **The hilum is a scar that marks the point where the seed was attached to the fruit wall by the funicle. Eg., Pea, bean, gram, nearly 82% plants have anatropous kind of ovule**

C-Hemi-anatropous/Hemitropous

The body of these ovules with micropyle and chalaza come in straight line at a right angle in relation to the funicle, so it looks like the ovule is lying on its side. Eg. Rannunculus



C

D-Campylotropous

The body of this type is bent and the alignment between the chalaza and micropyle is lost and they are close to funicle. The embryo sac is only slightly curved. Eg., Chenopodiacea, Caperidacea

D

E- Amphitropous

The body of the ovule is very much curved that the embryo sac and the ovule itself take the shape of a horseshoe. Eg., Lemna, poppy, Alisma

E

F-Circinotropous

The funicle in this case is especially long that it creates a nearly full circle around the ovule whose micropyle is ultimately pointing upwards. Eg. Cactaceae, cactus

F

- ✓ **However, these may be subdivided into additional types (“ana-” and “ortho-”) based on the orientation of the vasculature.**

- ✓ **An ana-amphitropous ovule is one in which a vascular strand curves, traversing from the base of funiculus to the chalazal region of the nucellus; the nucellus is bent sharply in the middle along both the lower and upper sides, often with differentiated cells (called a “basal body”) at the angle of the bend.**

- ✓ **An ana-campylotropous ovule is similar to the ana-amphitropous type in vasculature, differing in that the nucellus is bent only along the lower side, with no “basal body.”**

- ✓ **An ortho-amphitropous ovule is one in which the vasculature is straight, leading from the funiculus base to the middle of the nucellus; the nucellus is bent sharply in the middle along both the lower and upper sides, often with a “basal body” present.**

- ✓ **An ortho-campylotropous ovule is similar to that of the ortho-amphitropous type, except that the nucellar body is bent only along the lower side, with no “basal body.”**

Functions of Ovules

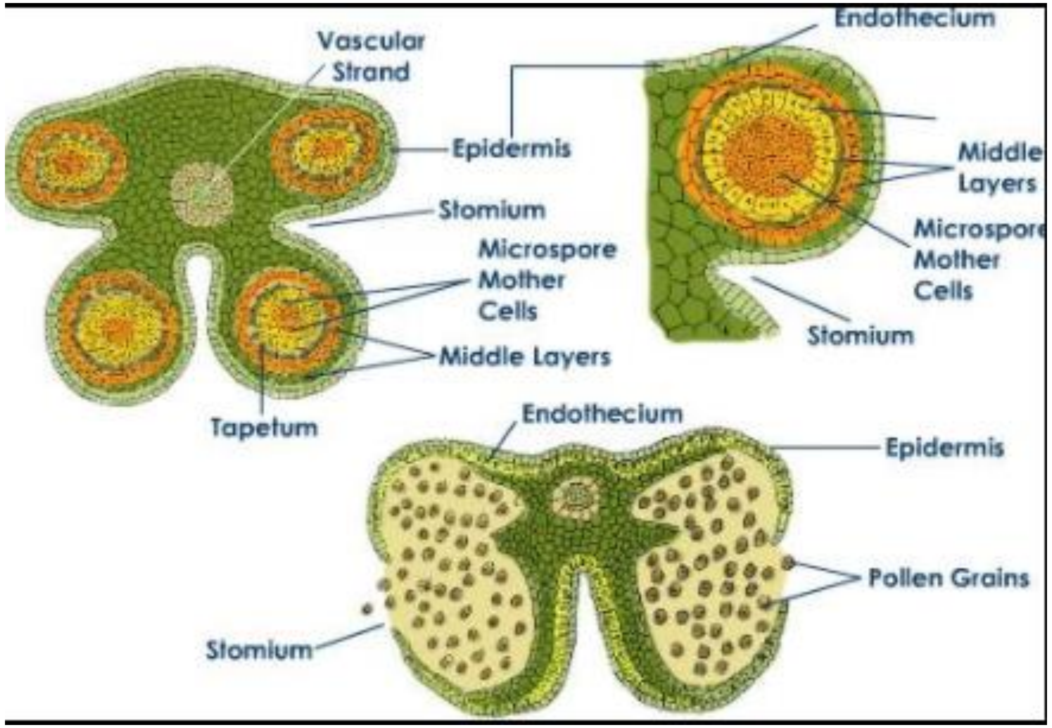
- **The ovule plays a vital role in sexual reproduction. Once a pollen grain lands on the stigma of a flower of its same species, it sends out a pollen tube down through the style.**

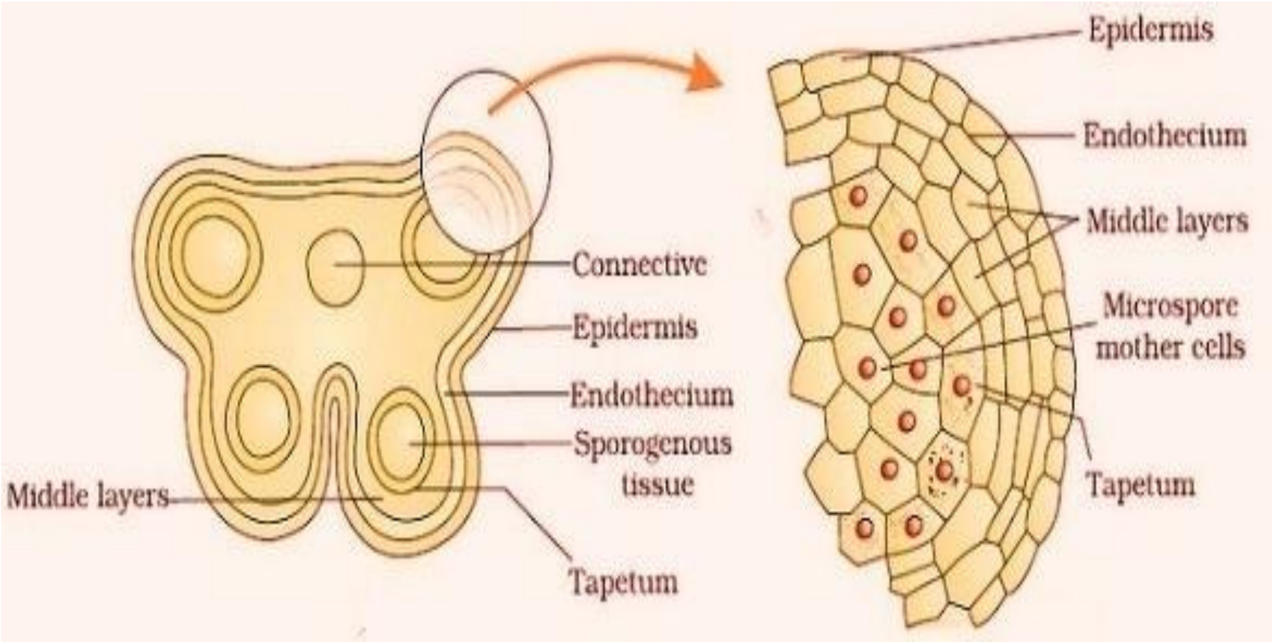
- **This tube then enters the ovary and reaches the ovule of the plant. Once that occurs, fertilization can arise as the nucleus of the pollen grain is sent down the tube to merge with the nucleus in the embryo sac.**

- **Note that the male alternative to the ovule is pollen, which contains the male gametophytes.**

Pollen development and structure

•Male reproductive part of a plant consists of an anther and the filament. Anther is bilobed containing 4 microsporangia in angiosperms so it is known as dithecous anther. Each microsporangium contains spores that produce male gametes. It has following layers:The outermost layer is known as the epidermis.
•Inner to the epidermis is the layer of endothecium.
•Inner to endothecium, there are 2-3 cell middle layers that usually disintegrate during anther maturation.
•The innermost layer is known as tapetum.
•Inner to the tapetum are the microspore mother cells which give rise to pollen grains through meiotic division.





ANTHER AND POLLEN DEVELOPMENT

- **ANTHER WALL DEVELOPMENT A cross-section of an anther reveals a division between the internal microsporangium, the cells of which undergo meiosis, and an outer anther wall.**
- **The development of the anther wall has provided some useful embryological features. A mature anther wall consists of few to several layers of cells.**
- **The outermost cell layer (just inside the epidermis) is termed the endothecium, which typically consists of enlarged cells with secondary wall thickenings functioning in anther dehiscence. The places from where dehiscence occurs, there endothecium cells are thin walled and are called as stomium. Below endothecium, 1-5 layered celled are of middle layer and inner most cells are tapetum.**
- **The secondary wall thickenings function by providing tensile force that pulls back the anther walls from the line or region of dehiscence.**
- **The innermost cell layer is termed the tapetum, which consists of metabolically active cells that function in the development of pollen grains.**

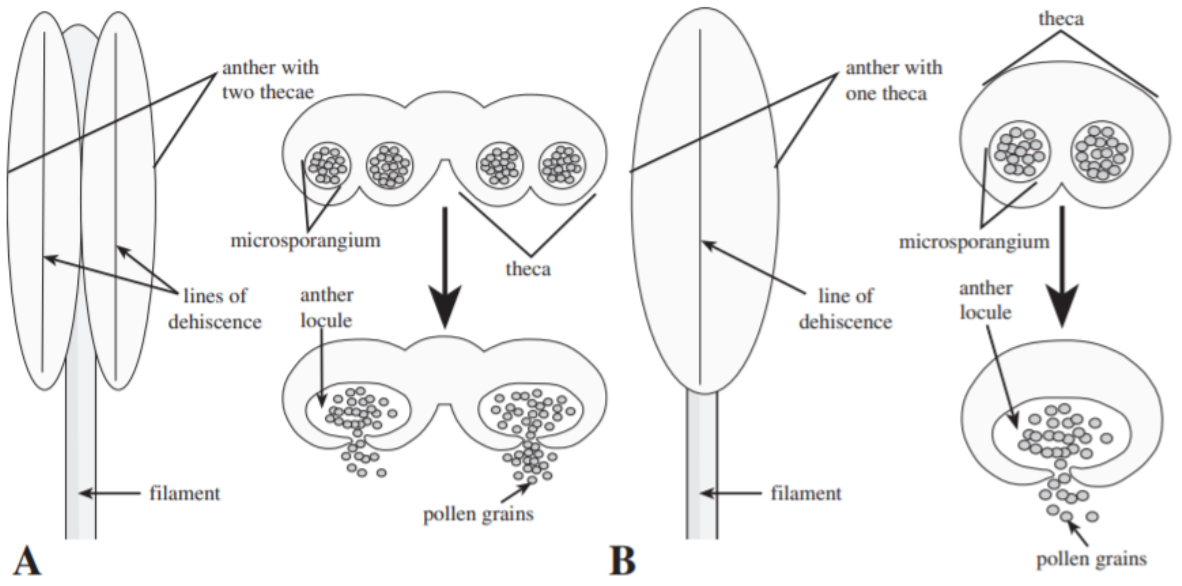


FIGURE 11.1 Anther types in the angiosperms. **A.** Dithecal. **B.** Monothecal.

Additional wall layers, termed middle layers, may occur between the endothecium and tapetum. Both the total number of wall layers and their developmental origin define various anther wall types.

Middle layers are soon lost as they provide nutrition to developing microspores. Tapetum cells become big, elongated and due to endomitosis they are polyploid and later provide nutrition to developing microspores.

- Early in development an anther contains two layers of cells, an outer epidermis and an inner layer of primary parietal cells.

- Cells of the primary parietal layer divide tangentially (parallel to the outer surface) to give rise to two layers of cells, secondary parietal cells.

- Based on the derivation of cell lineages, four general types of anther wall development have been defined:
- (1) basic, in which both secondary parietal cell layers divide to yield two middle layers;
- (2) dicotyledonous, in which only the outer secondary parietal cell layer divides to yield the endothecium and a single middle layer;
- (3) monocotyledonous, in which only the inner secondary parietal cell layer divides to yield the tapetum and a single middle layer; and
- (4) reduced, in which the secondary parietal cells do not divide further and develop directly into the endothecium and tapetum, respectively

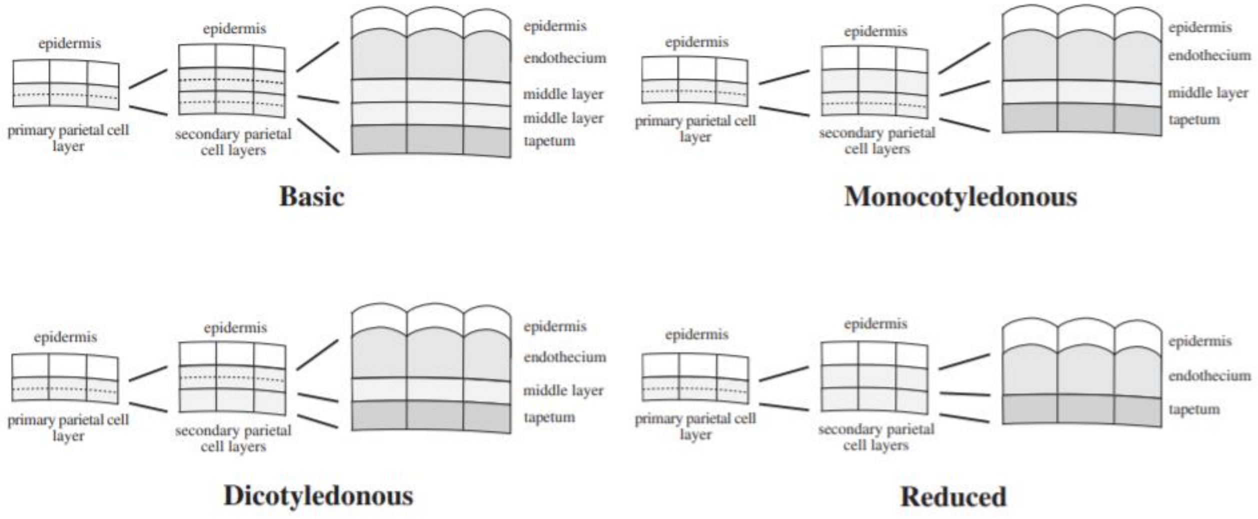


FIGURE 11.2 Anther wall development, outer epidermis at top.

- Another embryological character concerns the development of the tapetum, with two basic types defined.

- In some angiosperms the tapetum remains intact with no breakdown of cell walls. This tapetal type is called secretory (or glandular; Figure 11.4A,B) because of the implication that compounds are secreted into the locule of the anther that function in pollen development.

- In other angiosperm taxa the tapetal cell walls break down, with release of the cytoplasm of the tapetal cells into the locule. This latter tapetal type is called amoeboid (plasmodial or periplasmodial; Figure 11.4C,D) because the cytoplasmic contents surround developing pollen grains like an amoeba surrounds food.

- Subtypes of the secretory and amoeboid tapetal types have been proposed by some, based on fine developmental differences.

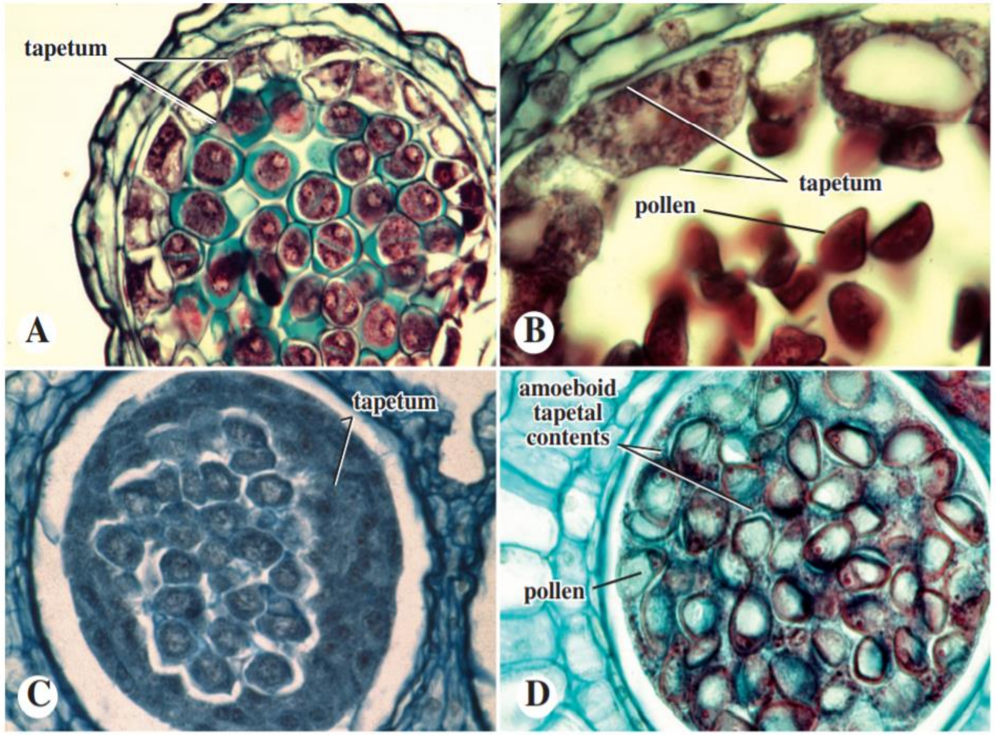


FIGURE 11.4 Anther cross-sections, showing different tapetum types. **A,B.** Secretory tapetum, the cells of which remain intact during pollen development (*Lophiola aurea*). **C,D.** Amoeboid tapetum, in which the cells break down, releasing their cytoplasmic contents into the anther locule. Early stage at left, later stage at right for both types (*Lachnanthes caroliniana*).

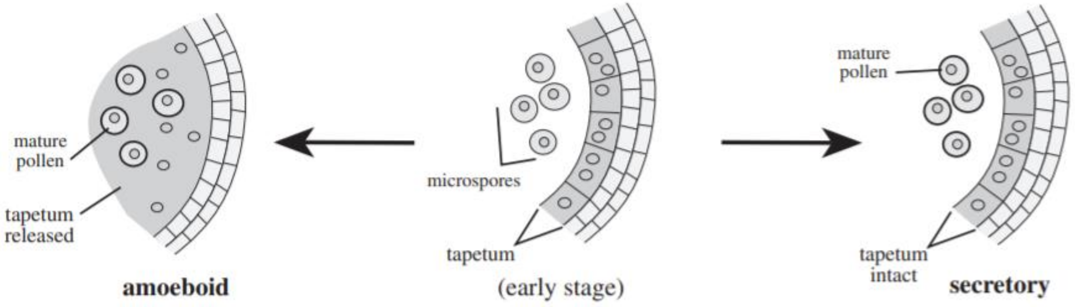
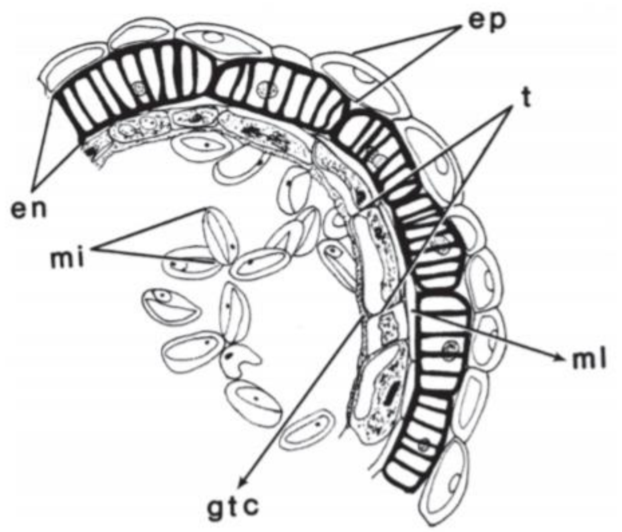


FIGURE 11.3 Tapetum types.

- **A final embryological character dealing with the anther wall is endothecial anatomy.**

- **Two basic types of endothecial cells have been defined based on the structure of the secondary wall thickenings.**

- **A girdling endothecium is one in which the secondary wall thickenings form rings with cross bridges between them. A spiral endothecium is one in which the secondary wall thickenings are spiral or helical in shape.**

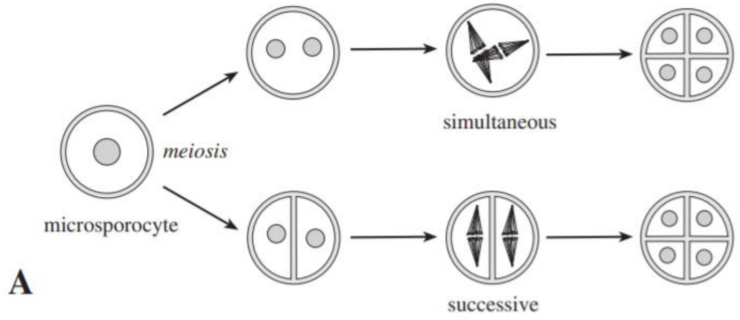


A girdling anther endothecium type. Symbols: en = endothecium; ep = epidermis; gtc = glandular tapetal cell; mi = microspore; ml = middle layer; t= tapetum.

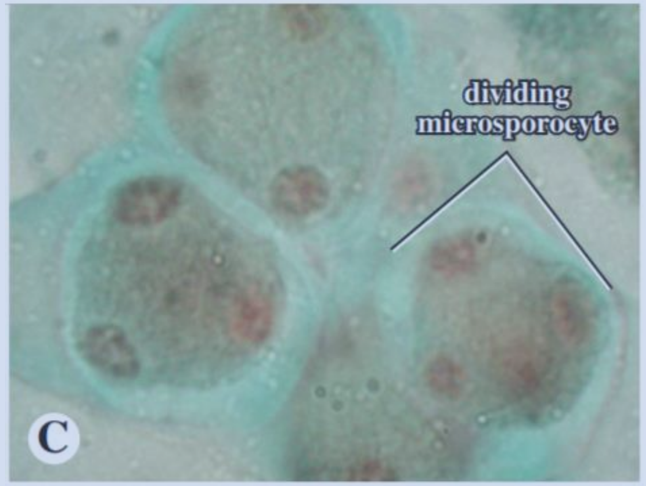
POLLEN DEVELOPMENT

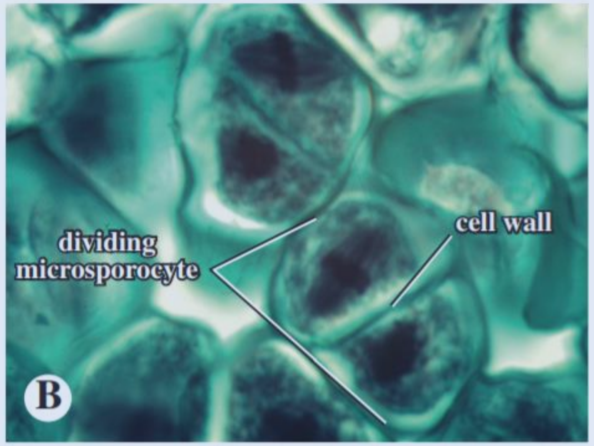
- **Development of microspores from microsporocytes is termed microsporogenesis.**

- **There are two basic types of microsporogenesis as determined by the timing of cytokinesis, which is the formation of a plasma membrane and cell wall that divides one cell into two (Figure A).**



- **If cytokinesis doesn’t occur until after meiosis II, then microsporogenesis is simultaneous (Figure C). Simultaneous microsporogenesis results in cell formation only after meiosis II.**





- **If cytokinesis occurs after meiosis I, then microsporogenesis is successive (Figure B). Successive microsporogenesis results in two cells after meiosis I and four cells after meiosis II.**

Microsporogenesis. A. Diagram showing two major types, simultaneous and successive. B. Successive microsporogenesis (Lophiola aurea). Microsporocyte at anaphase II of meiosis. Note that cytokinesis, resulting in cell wall formation, has occurred after meiosis I. C. Simultaneous microsporogenesis. Note lack of cell wall after anaphase II of meiosis.

- Development of pollen grains (male gametophytes) from microspores is called microgametogenesis, technically beginning with the first mitotic division of the single microspore nucleus.

- One embryological character concerning microgametogenesis is the number of nuclei present in the pollen grain at the time of anthesis, or flower maturation.

- Most angiosperms have pollen grains that are binucleate, containing one tube cell/nucleus and one generative cell/nucleus.

- The generative cell divides to form two sperm cells only after pollen tube formation. In many angiosperm taxa, however, the pollen at anthesis is trinucleate, caused by division of the generative cell prior to pollen release.

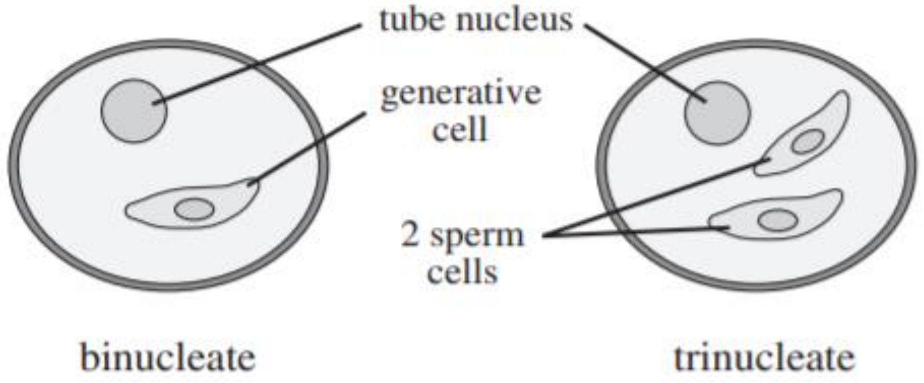


FIGURE 11.7 Microgametogenesis. Pollen nucleus number at anthesis.

Two major processes in sexual reproduction of seed plants, then, are pollination, the transfer of pollen grains from microsporangia to the ovule or stigma, and fertilization, union of sperm and egg. Many of the structural modifications of seed plants function in this transfer of pollen and the subsequent development and propagation of seeds.

In gymnosperms—cycads, Ginkgo, conifers, and Gnetales— pollen grains are almost entirely transported by wind. Because transport by wind is indirect, it necessitates the production of relatively large numbers of pollen grains to overcome the very low probability that any given pollen grain will make it to the ovule. In contrast, the great majority of angiosperms are animal (mostly insect) pollinated, which appears to be the ancestral condition for the family (Chapter 6), although wind pollination has arisen secondarily in several groups of flowering plants (see later discussion).

FLOWERING PLANTS Angiosperms have largely evolved very specialized floral structures that are adaptive in promoting animal pollination. Animal pollination is much more directed and precise, necessitating the synthesis of many fewer pollen grains to effect fertilization of the eggs within ovules. The basic adaptive “strategy” of animal-pollinated flowering plants has been the evolution of an attractant and a reward. The attractant works to entice the animal to the flower, either by vision or by odor. A visual attractant is usually a showy perianth (corolla and/or calyx) that may be brightly colored or otherwise contrasting with the external environment, e.g., a white perianth at night. Other floral parts, such as stamens (e.g., Hibiscus), staminodes (e.g., members of the Aizoaceae, Cannaceae, or Zingiberaceae), corona (e.g., Crinum, Narcissus, Passiflora), or even the gynoecium, may replace or augment the perianth as a visual attractant. Individual flowers may actually be small, but the accumulation of flowers in an inflorescence may provide a significant visual attractant. Olfactory attractants include the volatile compounds emitted by flowers, usually from the surface of the perianth. Most odoriferous flowers have a sweetish smell (e.g., Jasminum), but others emit compounds that mimic the smell of rotting flesh (e.g., Aristolochia, Arum, Stapelia).

Many species of flowering plants have evolved structures or exudates that act as a reward, ensuring that the animal pollinator will consistently return to transport pollen. The most common floral reward is nectar, a fluid primarily rich in sugars, secreted from specialized regions or organs of the flower called nectaries (Chapter 9). Nectaries are specialized tissues or organs that may be located within the gynoecium (e.g., the “septal” nectaries of many monocots), on the perianth, or at the base of and often surrounding the gynoecium or androecium. (Although nectar usually functions as a food source and reward for the prospective pollinator, some nectaries are “extra-floral” and may function as a reward for insects, such as ants, that protect the plant from herbivory by other animals.) Another pollination reward is pollen itself, which is a relatively rich source of protein. Some flowering plants produce waxes (e.g., Krameria) or resins (e.g., Clusia) as a reward. Finally, in some rare cases, insects may obtain specific chemical compounds that are used to attract a mate. Although the general strategy of pollination in most flowering plants is to provide a reward (thus, presumably, increasing the fitness of both plant and animal), not all animal-pollinated flowers do this. Some flowers have evolved structures or mechanisms to “trick” the animal to transport pollen, possibly with an adverse affect on the reproductive success of the animal. For example, in certain water lilies and orchids, the nectar may actually function to trap or even drown the insect to promote pollination. Other species of orchids actually mimic (by sight and odor) the female of an insect (usually a wasp), fooling the male to attempt to copulate with the flower, which, in the process, transports pollen.

POLLINATION MECHANISMS

Many, if not most, species of angiosperms have evolved specialized pollination mechanisms in which structural modifications are correlated with a specific agent of transferring pollen. Knowledge of the pollination agent can give insight into the function, homology, and evolution of associated floral features. The following are a summary of these general correlations or "syndromes."

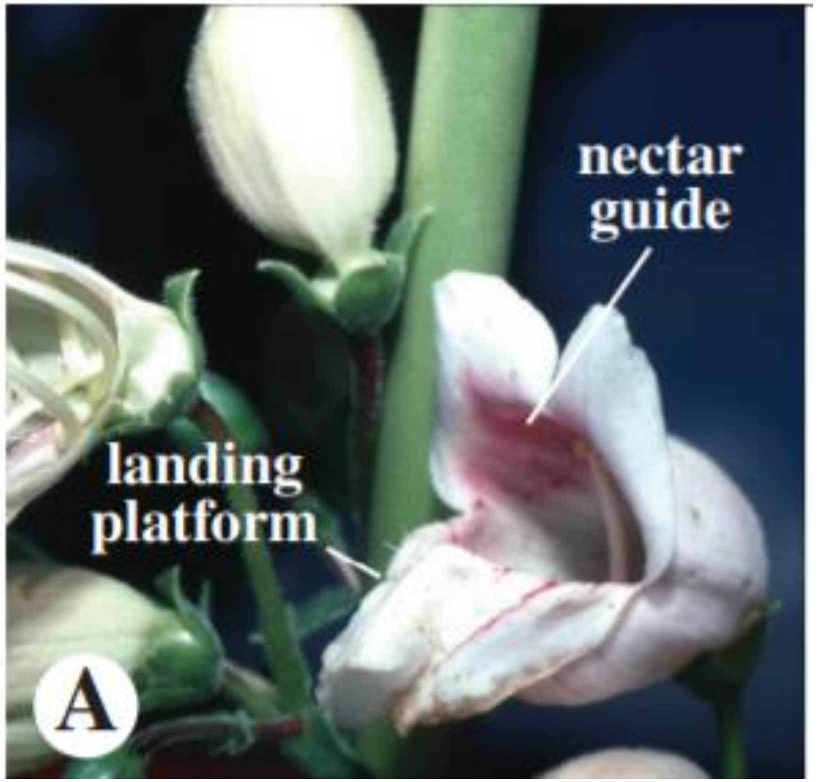
- **Insect pollination (or entomophily) is undoubtedly the most common type in angiosperms.**

- **Bee pollination (melittophily or hymenopterophily) is correlated with flowers that tend to be showy, colorful, and fragrant.**

- **The flowers often have specialized color patterns called nectar guides, which function to attract and orient the bee to maximally effect pollination.**

- **In many bee pollinated flowers, nectar guides may be correlated with the anterior perianth part(s) (usually petals or corolla lobes) modified as landing platforms (Figure A), on which the bee lands to more efficiently gather nectar or pollen and more effectively cause pollination.**

- **Ant pollination (myrmecophily) occurs more often with flowers that are low growing and inconspicuous.**



flowering plants maintain various populations of undifferentiated stem cells mostly in meristems. Meristematic tissue is capable of growth and differentiation to form vegetative tissues and organs, eventually giving rise to reproductive organs containing diploid sporogenous cells. A strict male germline is only established after meiosis when haploid microspores divide asymmetrically to form a small germ cell

The most widespread group of pigmented flavonoids is the anthocyanins, which are responsible for most of the red, pink, purple, and blue colors observed in plant parts. By coloring flowers and fruits, the anthocyanins are vitally important in attracting animals for pollination and seed dispersal. Anthocyanins are glycosides that have sugars at position 3 (Figure 13.13B) and sometimes elsewhere. Without their sugars, anthocyanins are known as anthocyanidins (Figure 13.13A). Anthocyanin color is influenced by many factors, including the number of hydroxyl and methoxyl groups in ring B of the anthocyanidin (see Figure 13.13A), the presence of aromatic acids esterified to the main skeleton, and the pH of the cell vacuole in which these compounds are stored.

Auxin Promotes Fruit Development Much evidence suggests that auxin is involved in the regulation of fruit development. Auxin is produced in pollen and in the endosperm and the embryo of developing seeds, and the initial stimulus for fruit growth may result from pollination. Successful pollination initiates ovule growth, which is known as fruit set. After fertilization, fruit growth may depend on auxin produced in developing seeds. The endosperm may contribute auxin during the initial stage of fruit growth, and the developing embryo may take over as the main auxin source during the later stages. Figure 19.39 shows the influence of auxin produced by the achenes of strawberry on the growth of the receptacle of strawberry.