EMBRYOLOGY OF BRUNELLA VULGARIS L.1

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ABSTRACT

Lae a ther development conforms to the dicotyledonous type. The tapetum is glandular and dimorphic. The pollen grains are tetrahedral, isobilateral and decussate and shed at two celled stage. The development of the megagametophyte is according to the Polygonum type. Obturator and hypostase are present. The endosperm is *ab initio* cellular and its development conforms "Brunella type". The embryo development follows the Mentha variation of Onagrad type. The seed coat is 1-3 loyered The pericarp consists of three zones. The outer epidermis is mucilaginous. At maturation, the hypodermal layer of inner epidermis forms the main protective layer.

INTRODUCTION

Brunella vulgaris L. belongs the sub-tribe Scutellarieae, and the tribe Stachydeae of the family Lamiaceae. Schnarf (1931) and Davis (1966) has reviewed the earlier embryological work on the family. Casual work on embryology of certain members of the tribe Stachydeae has been done by Ganguly (1948), Jaitly (1966), Santha Kumari (1976), Tiagi (1969), Dwivedi and Joshi (1980) and Dwivedi (1981). The present study deals with the embryology and seed coat and pericarp structure of Brunella vulgaris 1.

MATERIALS AND METHOOS

The materials young flower buds, open flowers, fruits and nutlets of all possible age were collected from Darjeeling and fixed in formalin-acetic-alcohol. Customary methods of dehydration, infilteration and embedding were followed, sections cut at a thickness of 5-10 microns were stained with iron alum-haematoxylin using safranin as counter stain.

OBSERVATIONS

Microsporogenesis and male gametophyte

In transection the young anther is four lobed and has a vascular supply in the centre (Fig. 1.). The anther develops according to the Dicotyledonous type (See Davis, 1966) (Figs. 2-5). The tapetal cells are glandular, binucleate and dimorphic (Figs. 5,6). The endothecium develops characteristic fibrous thickenings at binucleate pollen grain stage (Fig. 13). The meiotic divisions of pollen in mother cells are of simutaneous type. Tetrahedral (Figs. 9, 10), decussate (Fig. 11) and isobilatetral (Fig. 12) types of microspore tetrads are formed. The pollen grains are hexaporate and shed at two celled stage (Fig. 14).

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Figs. 1-27. Sporogenesis and development of gametophytes in Brunella vulgaris L. Fig. 1-14. Microspohogeness and development of male gametophyte. Fig. 1. cross section of a young anther. Fig. 2. T. S. of anther lobe showing archesporial cells. Figs. 3, 4. Portions of anther lobes showings differentiation of different layers of anther. Fig. 5. Portion of anther lobe showing 2-nucleate tapetum cells and pollen mother cells, Fig. 6. T. S. anther lobe showing tetrads. Fig. 7. T. S. of anther showing pollen grains and degenerating tapetum. Fig. 8. T. S. of mature anther, Fig. 9, 10. Tetrahedral, Fig. 11. Opposite decusate. Fig. 12. Isobilateral type of microspore tetrads. Fig. 13. Portion of anther lobe showing binucleate pollen grain and endothecium thickening. Fig. 14. Binucleate pollen grain. Figs. 15-27. Meg.sporogenesis and the development of female gametophyte. Fig. 19. L.S. of ovule showing meg.spore linear tetrad and hypostase. Fig. 22. L. S. of ovule showing meg.spore linear tetrad and hypostase.

(Ant-Antipodal cells; Ar-Archesporial cells; C-tpc-connective tapetall cell; cut-Cuticle; E-Epidermis; Eg-Egg; Ent:-Entothelium; Eth: Endothecium; Hy-Hypostase; Ml-Middle layer; Mmc-Megaspore mother cell; Ne-Nucellar epidermis; ob-Obturator; Pn--Polar nuclcus; Ppl-Primary parietal layer; Psl-sporogenous layer; Syn-Synergids; Tpc-Tapetal cell; Tpl-Tapetal layer; Vs-vascular supply.



Figs. 28-49. Endosperm, embryo, seed coat and pericarp development in Brunella vulgaris L. Figs. 28-33. Stages in the development of endosperm. Note the degenerated synergids, pollen tube, hypostase and endosperm. Figs. 2 34-44. stages in the development of embryo. For explanation see text. Figs, 45-49. Seed coat and pericarp development. Fig. 48. Portion marked 'X' in fig. 49 enlarged to show seed coat and pericarp structure. Fig. 49. L. S. of mature mericarp. Note degenerated micropylar haustorium and embryo.

(Ge-chalazal chamber; ch-chalazal haustorium Gut-cuticle; Dant-degenerating antipodal cells; Dmh-degenerating micropylar haustorium; Dsyn-degenerating synergids; En-endocarp; Ent : endothecium Ep-epicarp: I-integument; Mc-micropylar chamber; Mc-mesocarp; Mh-micropylar haustorial nucleus; Minc-megaspore mother cells; ob-obturator; Muc-mucilaginous cells Pn-polar nucleus; Ppl-primary parietal layer; pr-pericarp; Psl-primary sporogenous layer; Pt-pollen tube; Z-Zygote).

Megasporogenesis and female gametophyte:

The ovular primoridia differentiate as out growths from the placenta and develop into anatropous, unitegmic tenuinucellar ovules (Figs. 15-17). The cells of the funiculus near the micropyle become densely protoplasmic and enlarge to form an obturator (Figs. 22, 23).

A hypodermal archesporial cell functions directly as the megaspore mother cell (Figs. 15-17). The megaspore mother cell divides meiotically and produces a linear tetrad of megaspores (Figs. 18, 19). The three micropylar megaspores degenerate and the lower most functions (Fig. 20). The functional megaspore produces an eight nucleate polygonum type of embsyo sac by three successive nuclear divisions (Figs. 21, 23-27). The mature embsyo sac is comparatively broader in the micropylar part and narrow in the chalazal region. An endothelium of richly protoplamic cells surrounds the narrow part of the embryo sac (Fig. 27).

At the stage of linear tetrad of megaspores a hypostage is formed by the cells underlying the chalazal end of the gametophyte along with nucellar cells. They show sclerified thickenings. The hypostage persists upto the late stage of endosperm development (Figs. 18-22 & 24-33).

Endosperm

The primary endosperm nucleus lies in the centre of embryo sac (Fig. 28). At the time of division it moves toward the chalazal region, there it divides by transverse wall to produce a small chalazal and a large micropylar chambers (Figs. 29, 30). The nucleus of the chalazal chamber divides without any wall formation and organizes two nucleate chalazal chaustorium (Figs. 31-33). The chalazal haustorium shows lateral bending where it is completely encircled by the hypostage tissue (Figs. 28-33). At heart shaped embryo stage it gets empty (Fig. 43):

The nucleus in the micropylar chamber divides first vertically (Fig. 31) and then transeversely so as to form two tiers of two cells each (Fig. 32). The upper tier constitutes micropylar haustorium and the lower middle *teir contributes toward the formation* of endosperm proper by an isthmus (Fig. 33). The micropylar haustorium degenerates and devoid of cell contents at the heart-shaped stage of the embryo (Fig. 43).

The cells of lower middle chamber undergo repeated transverse and vertical divisions so as to organize a massive cellular endosperm proper (Figs. 33, 43 & 49). The mature seed is endospermic. The entire gamut of endosperm development conforms to "Brunella type" according to schnarff (1917).

Development of Embryo:

The development of the embryo takes place when a fair amount of endosperm tissue is formed. The zygote elongates extensively forming a long tubular process to go deep inside the endosperm (Figs. 33). The first divsion of the zygote is followed by a transverse wall forming a bi-celled proembryo (Fig. 35). The apical cell ca undergoes a vertical division giving rise to two juxtaposed cells and the basal cell cb divides transversely forming two superposed celss m and ci (Fig.36). The lowermost cell of the tetrad, ci undergoes transverse division forming two superposed cells, n and n^1 (Fig. 37).

In the meantime the daughter cells of ca divides by another vertical division at right angle to previous one forming quadrant q (Fig. 36). A transverse division of the quadrant results in an octant, arranged in two tiers $(1, 1^1)$ of four cells each (Fig. 37). Periclinal division in these tiers delimits a single layered dermatogen de (Figs. 39, 40). The inner group of cells of the tiers 1 and 1^1 after another periclinal divsion produce periblem pe and plerome Pl (Figs. 39, 40). By further divisions the cells of tier 1 differentiate into the stem tip (Pvt) and region (pco) (Fig. (4). Whereas the cells of I^1 tier give rise to the hypocotyledonary part (Phy) of the proembryo, which finally forms the hypocotyl-radicle axis (Figs. 42, 44). The cell m divides transversely to produce two daughter cells d, f (Fig. 39). The cell *d* directly functions as the hypophysis and undergoes transverse and vertical divisions to produce inititials of the root cortex iec and central cylinder of root (icc) (Fig. 40). These initials by further divisions contributes in the formation of the root cortex and root cap respectively. The cells derived from f, n and n^1 constitute a 6-7 celled, uniseriate filamentous suspensor s, which degenerates when the proembryo attains maturity. Thus the embryo development conforms the Mentha variation of Onagrad type according to Johanson (1950) or period I, series A₂, Megarche type IV of souege (1951).

Development of Seed Goat and Pericarp:

All the four ovules never develop into the seeds, oftenly one or two ovules become abortive. A young ovule consists of 5-6 layered integument at the megaspore mother cell stage (Fig. 45). The integument (seed coat) comprises 7 -10 layered structure at the time of endosperm development (Figs. 46, 47). In a fully matured fruit (mericarp) the seed coat is formed of outer epidermis and 2 hypodermal layers and the rest of the cell layers break down and finally consumed during the development of the embryo. The cells of the outer layer of the endosperm develop thickening on their inner tangential and radial walls (Fig. 48).

In pericarp, the hypodermal layer of the inner epidermis becomes lignified as a protective layer. The middle layer cells are empty and thin walled. The outer epidermal layer is mucilagenous and covered with a thick cuticle (Figs. 48).

DISCUSSION

The development of anther wall corresponds to the Dicotyledonous type (Davis, 1966) in all the members including the present species. The tapetum is of secretory type. Endothecial fibrillar thicknings is absent in *Leucas aspera* (Tiagi, 1979).

A single hypodermal archesporial cell is observed in the ovular primordium in the present study, has also been described in *Anisomeles indica* by Jaitly (1966).

The endosperm in **B**. vulgaris is ab initio cellular of "Brunella type" according to Schnarf (1917). The chalazal haustorium is coenocytic and binucleate. The bicelled, uninucleate chalazal haustorium has been observed in Scutellaria gulariculate (Schnarf, 1917) Anisomeles indica (Ganguly, 1948 and Jaitly, 1966); Scutellaria violacea and Westringia rigida (Santha Kumari 1976). Dwivedi and Joshi (1980) have observed 4-celled uninucleate chalazal haustorium in Craniotome versicolor.

In Anisomeles indica the micropylar haustorium is 4-celled (Ganguly, 1948 and Jaitly, 1966). A multinucleate (upto 12 nuclei) micropylar haustorium has been observed in Leucas aspera (Murthy, 1941; Jaitly, 1967 and Tiagi, 1979) and in Leonurus sibiricus (Ganguly 1948). Dwivedi and Joshi (1980) has reported 4-celled uninucleate micropylar haustorium in Graniotome versicolor.

The development of embryo in Brunella vulgaris is of Onagrad type but is of capsella-bursa-pastoris type in Leonurus sibiricus (Ganguly, 1948) and of Solaned type in Leucas mollissima (Tiagi, 1979).

In the present taxon seed coat is 1-3 layered. Two layered seed coat has been reported, in Anisomeles indica (Ganguly, 1948)

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