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# Studies in the Family Saxifragaceae VI. Structure and Development of Gametophyte in Deutzia corymbosa R. Br. and Dichroa febrifuga Lour.

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The earlier work, which was treated under the family *Hydrangeaceae* (Davis, 1966) shows that very little is known about the embryology of this interesting group. Subsequently, no additional information has become available. In the present paper, the structure and development of the gametophytes are described for *Deutzia corymbosa* (Tribe Philadelpheae) and *Dichroa febrifuga* (Tribe Hydrangeae), both belonging to the sub-family Hydrangeoideae (Engler, 1930).

## Material and Method

The material for the present study was collected by Professor V. Puri from Nepal and was kindly made available to me for study. Preserved samples have been maintained at the Department of Botany, Meerut College, Meerut (India). After following the usual methods of dehydration, clearing and embedding, the material was sectioned at  $10-12 \mu$  thickness. The slides were stained with safranin-fast green and iron alum-haematoxylin-fast green combinations which gave satisfactory results.

## **Observations and Discussion**

Microsporangium and male gametophyte: The microsporangial wall consists of an outer epidermis, single layered endothecium, 1 or 2 (D.corymbosa) or 2-4 (D.febri-fuga) middle layers and an innermost tapetum (Fig. 1, 2). The tapetum is secretory in nature and in D.corymbosa its cells become binucleate at maturity. In Astilbe rivularis (Webb, 1902) and Saxifraga diversifolia (Saxena, 1964a) in contrast, they remain uninucleate. Before dehiscence of the anther, characteristic fibrous thickenings appear in the cells of endothecium. By this time the tapetum degenerates in D.corymbosa but persists in D.febrifuga (Fig. 1-3).

Different stages of microsporogenesis could not be observed but in *D. corymbosa* tetrahedral tetrads and occasionally decussate tetrads of microspores have been observed (Fig. 4–7, 12). In *D. corymbosa* the microgametogenesis may progress while



Plate I

the microspores are still arranged in tetrads (Fig. 4–6). As a result of unequal division, two cells are formed; a small generative and a comparatively larger vegetative one (Fig. 8–11, 13). The two nuclei may sometimes be almost equal in size (Fig. 10). A few pollen grains were found to have a nucleus and an arc-like degenerating mass (Fig. 11). In one case, one of the nuclei of the pollen grain was binucleolate (Fig. 9). In both the species investigated at present, as well as in *Astilbe japonica* (Webb, 1902), *Parnassia palustris* (Pace, 1918), *Vahlia viscosa* (Raghavan and Srinivasan, 1942), *Parnassia nubicola* (Sharma, 1968), *Bergenia ciliata* and *Mitella diphylla* (Saxena, 1969, 1971) the pollen is shed at the 2-celled stage, whereas in *Saxifraga diversifolia* (Saxena, 1964a) it is shed at the 3-celled stage. A large number of sterile pollen grains have also been observed in *D.febrifuga*.

Megasporogenesis and Female Gametophyte: The ovules are anatropous and unitegmic. Besides the Hydrangeoideae, unitegmic ovules have also been reported in Saxifraga foliosa (Harmsen, 1939), Saxifraga hieracifolia (Wiggins, 1959), Peltiphyllum, Kirengeshoma and Escallonia (see Davis, 1966). The cells forming the nucellar epidermis are larger in size as compared to those in the neighbouring integumentary cells. A hypodermal archesporial cell differentiates in the nucellus of D. febrifuga (Fig. 14). However, in *D. corymbosa* the earliest stage observed was a tetrad of megaspores. The archesporial cell directly functions as a megaspore mother cell without cutting out any parietal cell and as such the ovules are tenuinucellate. The megaspore mother cell divides to form dyads (Fig. 15, 16). The upper dyad may degenerate sometimes without undergoing further division (Fig. 17). The lower one subsequently divide to form two megaspores (Fig. 18, 19). Early degeneration of the two micropylar megaspores was observed in D. corymbosa where any megaspore is functional but more frequently the chalazal one (Fig. 20). Various types of megaspore tetrads have been observed in Saxifraga diversifolia and Parnassia nubicola (Saxena, 1964a, b). Rarely, more than one healthy megaspores of configuration "abCD" have been observed (Fig. 18). The functioning megaspore enlarges considerably and as a result the nucellar epidermis and other neighbouring cells of the nucellus are crushed. Even before the disappearance of nucellus, the cells of the integument surrounding the megaspores on the lateral sides become radially elongated and take a darker stain (Fig. 19). The integumentary tapetum, so differentiated, extends from the micropylar to the chalazal end of the embryo sac (Fig. 22, 24). The formation of the integumentary tapetum has also been observed in several other members of the family (see Pace, 1912; Mauritzon, 1933).

#### Plate I

Figures 1, 12-18, 22-24: Dichroa febrifuga; Figures 2-11, 19-21, 25-30: Deutzia corymbosa (Fig. 1-3: Portions of the anther wall; Fig. 4-13: Pollen tetrads and stages of microgametogenesis, a decussate tetrad shown in Fig. 7; Fig. 14-30: Various stages of megasporogenesis and mega-gametogenesis, two healthy megaspores shown in Fig. 18, 19; Fig. 29: Showing division in the chalazal nuclei; Fig. 30: Showing division in the extruded micropylar nuclei)

d.m. degenerated megaspores; en. endothecium; ep. epidermis; f.t. fibrous thickening; i.t. integumentary tapetum; m.1. middle layers; n.e. nucellar epidermis; t. tapetum Scales: B for Figures 27, 28; A for the rest



## Plate II

Figures 31-36, 41: *Deutzia corymbosa;* Figures 37-40: *Dichroa febrifuga* (Fig. 31-38: Organisation of the embryo sac; areas enclosed in rectangles in Fig. 31, 33, 35 and 37 are enlarged in Fig. 32, 34, 36 and 38 respectively; Fig. 39: Organised embryo sac; Fig. 40: Egg apparatus enlarged; Fig. 41: Showing some supernumerary nuclei on the egg apparatus side)

a. antipodals; e. egg; i.t. integumentary tapetum; p. polar nuclei; s.n. secondary nucleus; s. synergid Scale: A for Figures 31, 33, 35; B for Figure 37; C for Figures 32, 34, 38, 40; D for Figures 36, 41; E for Figure 39

The nucleus of the functioning megaspore divides in two and subsequently in four (Fig. 20-28). In D. corymbosa the embryo sac elongates at the 4-nucleate stage and as a result of the enormous elongation, the micropylar portion of the embryo sac is pushed out of the ovule through the micropyle (Fig. 27, 28). The two extruded nuclei, embedded in a protoplasmic sheath, lie on the funicular side closely appressed to the ovule. The third division of megagametogenesis takes place immediately, i.e. when the micropylar pair of nuclei have passed out of the ovule (Fig. 29, 30, 37, 38). In D. febrifuga, in contrast, the embryo sac shows considerable elongation along the longitudinal axis of the ovule during the third division of megagametogenesis. But there the embryo sac does not protrude out of the ovule. The eight nuclei are organised into a Polygonum type of embryo sac. The egg apparatus in D. corymbosa is organised outside the ovule and lies in between the placenta and funiculus (Fig. 31-40). A similar condition has been observed in *Philadelphus* by Mauritzon (1933). However, in *Kirengeshoma* (Mauritzon, 1933) the micropylar portion of the embryo sac protrudes out of the endostome and eventually lies in the exostome. Extrusion of the micropylar portion of the embryo sac has also been observed in many other families and genera (see Maheshwari, 1950; Subramanyam, 1960; Johri, 1962; Masand and Kapil, 1966). Since the extruded part of the embryo sac serves as a micropylar haustorium, this sort of anomaly has been assigned a nutritive function.

It is interesting to note that the integumentary tapetum, which in the beginning extends from the micropylar to chalazal end, remains restricted to the chalazal region and extends only about half the length of the mature embryo sac (Fig. 38, 39). This condition may be the result of a sudden growth in the ovule on the micropylar side during the four-nucleate stage of the embryo sac. Fusion of the two polars may also take place outside the ovule in *D. corymbosa* (Fig. 32). In *D. febrifuga* the synergids are hooked and in both the plants the egg apparatus is devoid of any filiform apparatus. However, these structures have been observed in *Saxifraga hieracifolia* (Wiggins, 1959) and *Ribes cynosbati* (Saxena, 1970). In *D. corymbosa*, some supernumerary nuclei have also been seen on the micropylar end of the embryo sac before the organisation of the egg apparatus (Fig. 41).

#### Summary

The tapetum in both the species studied (*Deutzia corymbosa* and *Dichroa febrifuga*) is glandular in nature and the pollen is shed at the 2-celled stage. The ovules are anatropous, unitegmic and tenuinucellate. As a result of meiosis the megaspore mother cell develops into a linear tetrad. An integumentary tapetum develops after degeneration of the nucellus. Enlargement of the embryo sac is observed at the 4nucleate stage and in *D. corymbosa* the micropylar portion of the embryo sac protrude out of the ovule. The present observations show that the integumentary tapetum remains mainly restricted, at the later stages, only to the chalazal half of the ovule. The development of the embryo sac conforms to the Polygonum type.

#### Zusammenfassung

In beiden Arten (*Deutzia corymbosa* und *Dichroa febrifuga*) ist das Tapetum drüsig, und der Pollen wird im Zweizellstadium frei. Die Samenanlagen sind anatrop und haben ein Integument und einen dünnen Nucellus. Bei der Meiose entsteht aus der Makrosporenmutterzelle eine lineare Tetrade. Nach der Degeneration des Nucellus entsteht ein integumentäres Tapetum. Im Vierkernstadium vergrössert sich der Embryosack; bei *D. corymbosa* tritt der mikropyläre Teil des Embryosackes aus der Samenanlage aus. Das integumentäre Tapetum bleibt in den späteren Stadien im wesentlichen auf die der Chalaza anliegende Hälfte der Samenanlage beschränkt. Die Entwicklung des Embryosackes entspricht dem *Polygonum*-Typ.

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