J. Indian bot. Soc. **64** : 61—65, 1985.

EMBRYOLOGY AND SEED DEVELOPMENT IN *BEAUMONTIA GRANDIFLORA* WALL.

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ABSTRACT

Seed development in *Beaumontia grandiflora* Wall. has been invetsigated. Ovule is unitegminal, anatropous and tenuinucellar. Nucellus is thin and parenchymatous. The chalazal megaspore of the linear tetrad develops into an 8-nucleate embryo sac of the Polygonum type. Occasionally two lower megaspores of a tetrad are seen enlarged. The mature embryo sac contains starch grains. In few instances, two embryo sacs are reported in an ovule. Development of the endosperm is nuclear and the embryogeny follows the Solandad type. The seed coat is composed of 3-4 layers of cells with a prominent epidermis. Seeds are minute, dark brown and non-endospermic.

INTRODUCTION

The earlier literature on the embryology of Apocynaceae has been reviewed by Schnarf (1931) and Davis (1966). Although some work has been done on the structure and development of ovule and female gametophyte of a few taxa of the family (Andersson 1931, Meyer 1938, Rau 1940, Murty and Chauhan 1966, Maheswari Devi 1971, 1974 ; Bhasin 1971 and Lamba 1974), little attention has been paid to the development of the embryo, endosperm and seed coat. Periasamy (1963) has studied the development of seed in only three members of the family with special emphasis on the ruminate endosperm. The present investigation has, therefore, been undertaken to work out the developmental stages of a seed in *Beaumontia grandiflora* Wall.

MATERIAL AND METHODS

Young buds, flowers and seeds were fixed in Formalin-acetic-alcohol from local gardens and preserved in 70% alcohol. The material was dehydrated and infilterated through alcohol-xylol grades and embedded in paraffin wax of MP-58°C. A small surface of the embedded material was exposed by trimming with a razor blade. Then it was placed in a vial containing 15 ml. glacial acetic acid, 5 ml. glycerine and 80 ml. 65% ethyl alcohol at 25-27°C., for 2-3 days to facilitate the microtoming. Sections were cut at 7-15 microns and stained with safranin-light green combination.

OBSERVATIONS

Ovule—Numerous ovules are present in superior, bicarpellary and bilocular ovary. The ovule is anatropous, uniteg-

Accepted for publication on March 13, 1984.

The author is indebted to Prof. R. N. Kapil for his kind guidance and encouragement. Thanks are also due to Drs. N. K. Uberoi and Mr. A. C. Sood for certain facilities.

minal and tenuinucellar (Fig. 1). It originates on the thick placenta as a nucellar primordium and quickly grows in size. Considerable growth is noticed at the chalazal portion of the ovule after fertilization. Nucellus is thin and its cells appear parenchymatous. A few cells at the base of the nucellus in the chalazal region stain deeply due to the rich cytoplasmic contents. A single integument takes its origin early and deeply buries the nucellus. The integument is composed of 4-5 layers of cells towards the sides and 6-8 layers of cells at the basal region in its early stages of development (Fig. 1).

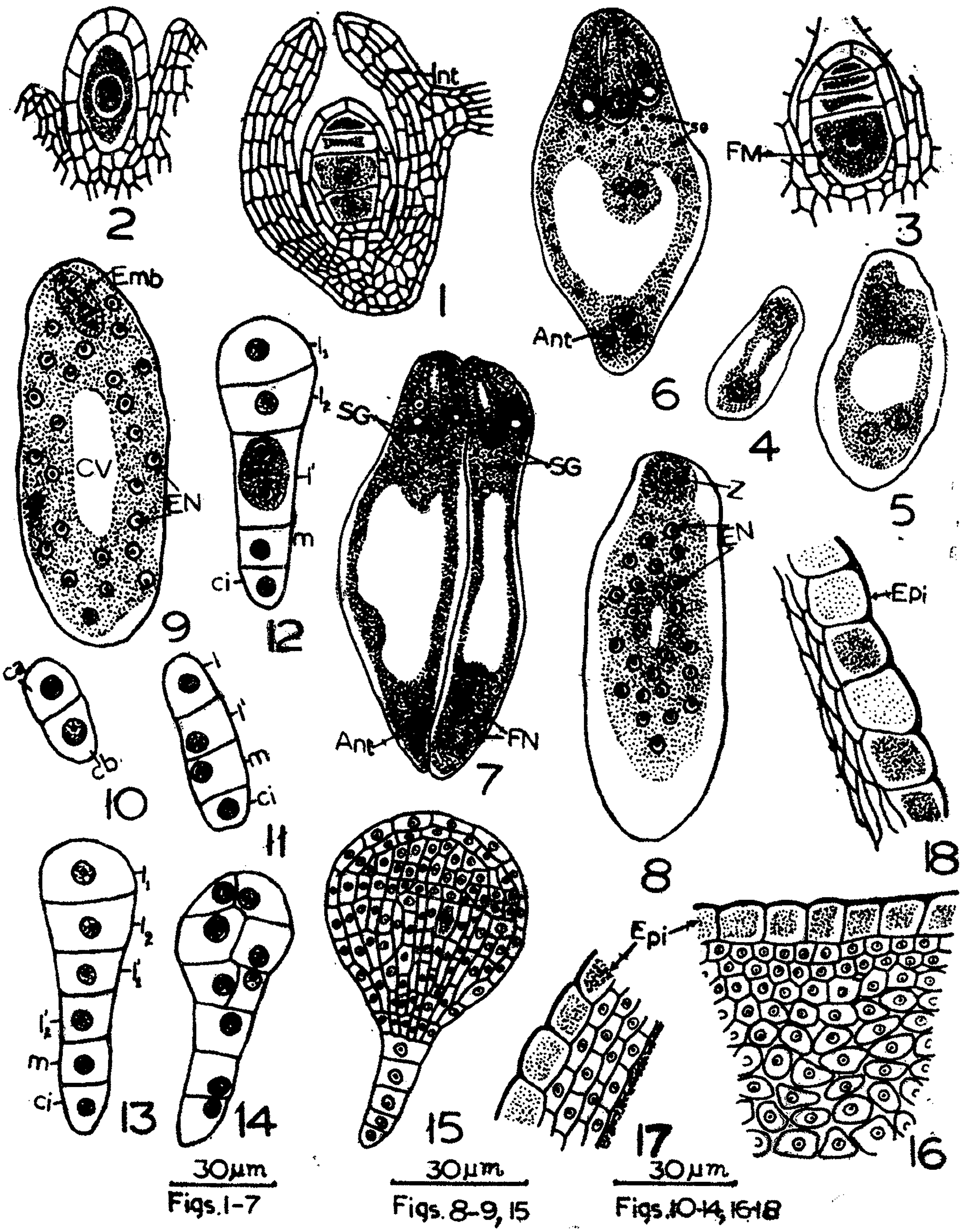
Megasporogenesis and megagametogenesis. The hypodermal archesporium is unicelled and directly functions as megaspore mother cell (Fig. 2). It enlarges and undergoes meiosis to produce four megaspores arranged in a linear fashion. The upper three megaspores degenerate, while the chalazal one remains functional (Fig. 3). Rarely the upper two megaspores of a linear tetrad disorganise and the lower two megaspores appear enlarged (Fig. 1). Nucleus of the functional megaspore divides thrice mitotically to form 2, 4, and 8-nucleate embryo sac (Figs. 4-6). Thus, embryo sac development is according to polygonum type. Nucellar epidermis begins disintegrating at the 2-nucleate stage of the embryo sac. An organised embryo sac consists of an egg-apparatus, two polar nuclei and three antipodal cells (Fig. 6). The synergids are pear-shaped with a prominent basal vacoule. Polar nuclei meet in the middle of the embryo sac and finally come to lie adjacent to egg. The uninucleate antipodals persist only till fertilization. The mature embryo sac is found full of starch grains. Rarely, two embryo sacs are recorded in an ovule (Fig. 7). Out of the two, one is the normal 8-nucleate embryo sac while the other embryo sac is abnormal because it shows one egg and one synergid at the micropylar end, and six free nuclei at the chalazal end.

Endosperm : Endosperm development is of the nuclear type. The primary endosperm nucleus moves to the centre of the embryo sac and divides repeatedly prior to the division of the zygote (Fig. 8). The nuclei are shifted to the periphery because of the formation of a central vacuole (Fig. 9). Cell formation in the endosperm occurs at the globular proembryo stage and usually starts from the periphery. Endosperm gets consumed by the growing embryo.

Embryo : The enlarged zygote under-

Figures 1-18. Stages in the development of seed in *Beaumontia grandiflora*. Figure 1. L. S. unitegmic, anatropous ovule, note 2-enlarged chalazal megspores. Figure 2. L. S. nucellus to show megaspore mother cell in meiotic prophase and developing integument. Figure 3. L. S. part of ovule showing functional chalazal megaspore. Figure 4. Bi-nucleate embryo sac. Figure 5. Four-nucleate embryo sac. Figure 6. Organised embryo sac with starch grains. Figure 7. Twin embryo sacs One normal and other abnormal). Figure 8. Free nuclear division of primary endosperm nucleus and initiation of central vacuole. Figure 9. Endosperm nuclei shifting towards periphery. Figure 10. two celled proembryo. Figure 11. Four celled proembryo. Figure 12. Proembryo showing division of the cell l′. Figure 13. Six celled proembryo. Figure 14. Globular proembryo. Figure 15. Globular embryo with well-developed suspensor. Figure 16. Ten-layered seed coat with prominent epidermal cells. Figure 17. Five-layered seed coat. Figure 18. Mature seed coat with persistent epidermis.

Ant, Antipodal cells; CV, Central Vacuole; Emb, Embryo; EN, Endosperm Nuclei; Epi, Epidermis; FM, Functional Megaspore; FN, Free Nuclei; Int, Integument; SG, Starch Grains; Z, Zygote.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goes a transverse division to produce two cells apical cell ca and basal cell cb (Fig. 10). The two cells divide further by a transverse division forming a row of 4 cells which may be designated asl, l′, m and ci (Fig. 11). Another transverse division in each of the two cells l and l′ results in the formation of a linear proembryo of 4-cells, l_1, l_2, l'_1 and l'_2 (Figs. 12, 13). Vertical and anticinal divisions in the three-cells l_1, l_2, and l'_1, give rise to a globular mass (Figs. 14, 15) from which arise the two cotyledons, the plumule and the root cap. Uniseriate suspensor of 4-6 cells is formed by the divisions in cells l'_2, m and ci (Fig. 15). The development of embryo conforms thus to the Solanad type.

Integument (*Seed coat*). The epidermal cells of the integument become enlarged at the 6-celled proembryo stage. These cells are quite prominent and persistant in the mature seed coat. Granular contents which stain dark red or purple with safranin, appear in a few cells of the epidermis. At the globular stage of embryo, the seed coat is 8-10 layered thick towards the sides (Fig. 16) and multilayered towards the micropylar and chalazal regions. The subsequent development of the embryo in the ovule results in the enlargement of the embryo sac thereby crushing the integumentary cells. Thus seed coat appears only 4-5 celled thick (Fig. 17) at the heart shaped stage of the embryo. By the time a well-defined embryo is formed, seed coat is only 3-4 layered thick showing a prominent epidermis and 2-3 layers of cells of integument (Fig. 18).

Seeds : Young seed is green and shiny while the mature seed is dark brown to black in colour. It is ovoid to oblong in shape with a diameter of 1.5-2.5 mm. Mature seed coat shows 3-4 layers of cells. Surrounded by the seed coat lies the well-developed embryo. Endosperm is absent.

DISCUSSION

In *Beaumontia grandiflora* the ovule is anatropous and nucellus is thin. The nucellar epidermis on the sides mingles with the integumentary tissue in *Rauvolfia canescens* (Meyer, 1938). The present study shows a well-defined nucellar epidermis which remains separate from the integumentary tissue. Such condition has also been recorded in the investigated members of the tribe Plumiereae (Maheswari Devi, 1971). As in *Voacanga grandiflora* and *Ervatamia heyneana* (Periasamy, 1963), the rapid growth is noticed at the chalazal portion of the ovule in *B. grandiflora* which is contrary to what is reported in *Tabernaemontana* sp. by Periasamy (1963).

Archesporium is single celled and hypodermal. It directly functions as megaspore mother cell which divides meiotically to form a linear tetrad as in other investigated members of Apocynaceae (Rau, 1940 ; Murty and Chauhan, 1966 ; Maheswari Devi, 1971), 1974, Bhasin, 1971 and Lamba, 1974). As in majority of the investigated taxa of the family, the present study also does not record any T-shaped tetrads. However, such feature has been reported in *Catharanthus roseus* (Maheswari Devi, 1971). According to Frye and Blodgett (1905). any of the megaspores of a tetrad may become functional in *Apocynum androsaemifolium* but in *B. grandiflora* the chalazal megaspore develops into 8-nucleate embryo sac of Polygonum type. However, the present investigation shows an occasional disorganisation of the upper two megaspores and the enlargement of the lower two ones which corroborates with the findings of Murty and Chauhan (1966) in *Lochnera pusilla*, and Lamba (1974) in *Rauvolfia serpentina*. Maheswari Devi (1971) recorded two embryo sacs in an ovule in *Rauwolfia tetraphylla*, *R. serpentina* and *Holarrhena antidysenterica*. The present work also shows the rare occurrence of twin embryo sacs in an ovule.

As reported in other members of the Family (Rau, 1940 ; Periasamy, 1963 ; Murty and Chauhan, 1966; Maheswari Devi, 1971, 1974 ; and Bhasin 1971) endosperm is nuclear ab initio but it becomes cellular in later stages. Attempts were made to trace out some stages of embryo development in *Lochnera rosea* and *Vinca minor* by Andersson (1931), and in *Cerbera odollam*, *Carissa carandas* and *Ichnocarpus frutescens* by Rau (1940). The present detailed investigation reports that the development of embryo in *B. grandiflora* follows the Solanad type. A similar condition has been recorded by Maheswari Devi (1974) in *Carissa spinarum*. As in *V. foetida* and *C. spinarum* (Maheswari Devi, 1971, 1974), a few epidermal cells of the seed coat in *B. grandiflora* contain granular brown contents. According to Maheswari Devi (1971) seed coat in *R. tetraphylla* and *V. foetida* is massive and many layeréd but in *C. roseus* and *Catharanthus pusillus*, all the layers of integument disintegrate and seed coat is constituted only by the outer epidemris. However, the seed coat in the present plant consists of 2-3 wall layers of the integument and an outer epidermis. Thus the embryology of *B. grandiflora* resembles closely to the other investigated taxa of the family Apocynaceae.

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