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Structure and Development of Seed Coat in *Cleome*

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The bitegmic, crassinucellate ovules are campylotropous in *Cleome monophylla*, *C. aspera*, *C. chelidonii*, *C. gynandra* and sub-campylotropous in *C. viscosa*. Ovular primordium is trizonate. Integuments are dermal in origin. Outer integument is non-multiplicative and remains two layered. Inner integument is multiplicative and becomes 7 - 10 layered. Epidermal cells of testa become thickened and develop tannin at the heart-shaped embryo stage. Tegmen at maturity differentiates into an outer fibrous layer (exotegmen) 5 to 6 layers of thin walled mesophyll cells (mesotegmen) and a lignified endotegmen. Mature seed is exotegmic, reniform, perispermous and endospermic. The origin and nature of the so called third seed coat is studied. Structure of mature seed coat provides a set of important characters for the identification of different species.

Key Words - Coat Endotegmen Epidermis Integument Ovule Seed Tegmen

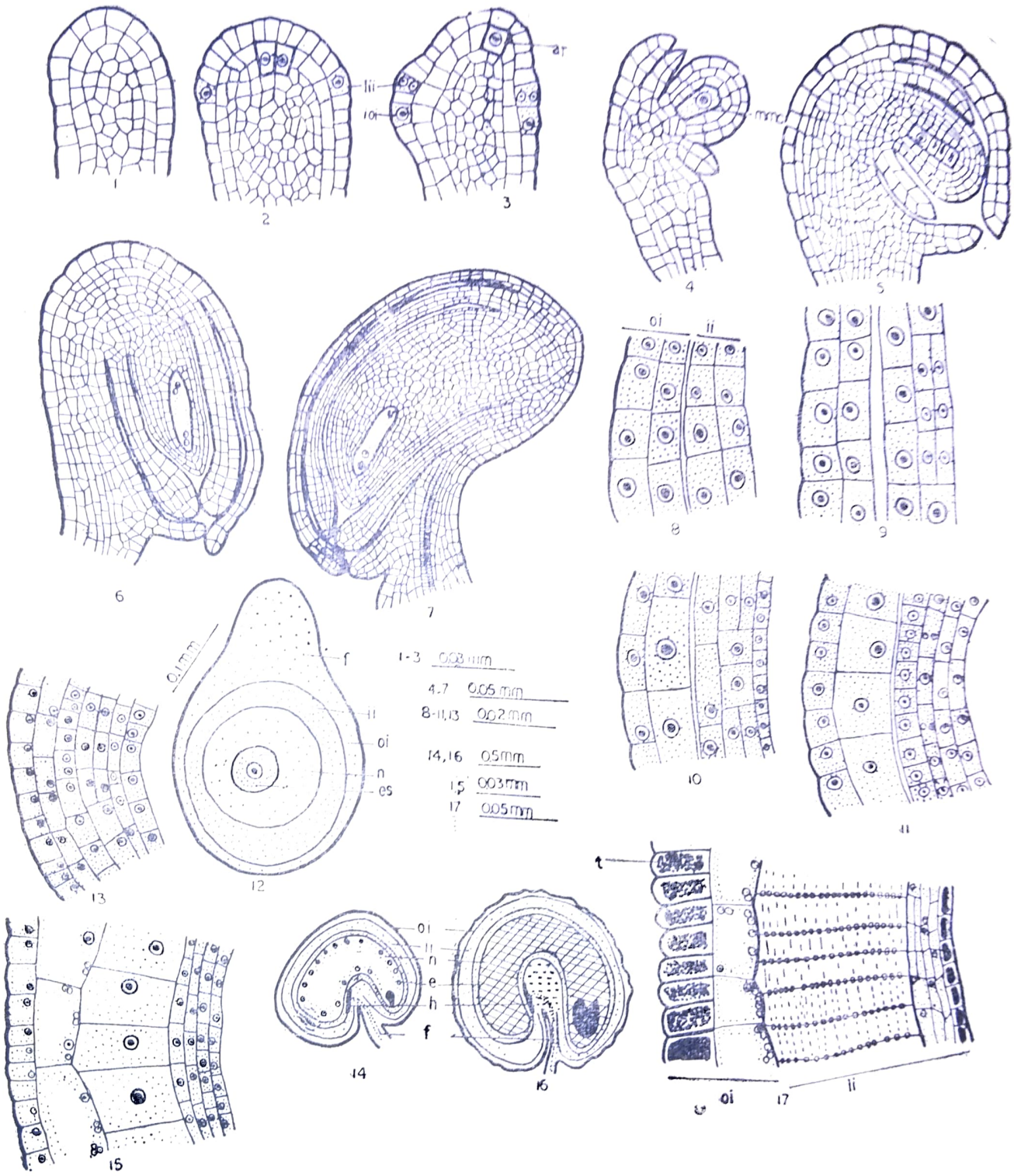
The importance of ovule and seed in the evaluation of the phylogeny of angiosperms and in solving certain taxonomic problems is stressed by Singh (1964), Wunderlich (1967), Corner (1976) and Bouman (1978). Bouman (1978) emphasised that complete ovular histogenesis is required when seed coat architecture is to be used for taxonomic purposes. The knowledge relating to the seed coat anatomy of Capparaceae is scanty and even the little work that is recorded has been described while dealing with its embryology (Raghavan, 1937; Narayana, 1962, 1965). Corner (1976) suggested that the seed coats in the family are complicated and require detailed study. In this paper detailed description of the ontogeny and anatomy of ovules and seeds in 5 species of *Cleome* i. e. *C. monophylla* Koeng., *C. chelidonii* L., *C. viscosa* L., *C. aspera* Koeng. and *C. gynandra* L. is presented.

MATERIALS & METHODS The material collected locally was fixed in FAA and customary methods of microtechnique were followed (Johansen, 1940). Sections were cut at thickness ranging from 6-15 μm and stained with Delafield's haematoxylin. The mature seeds were macerated following Jeffrey's technique (Johansen, 1940).

OBSERVATIONS The ovules are arranged in four rows on two parietal placentae in a bicarpellary unilocular ovary.

The trizonate ovular primordium is distinguished into three zones-an outer layer of dermatogen, a subdermal layer and an inner core which is bulky (Fig. 1). The primordium soon begins to curve due to greater mitotic activity of the basal cells on the convex or adaxial side of ovule.

The integuments are dermal in origin and the inner integument initiates earlier and is visible as a symmetrical ring by the time the archesporium is differentiated (Fig 2). The outer integument is asymmetrical and is initiated on the dorsal or convex side of the ovule in close proximity to the inner (Fig. 3). At a later stage, the outer integument although is visible on the funicular side it is poorly developed. As further development of both the integuments is identical a common account is given.The initials of the integument can readily be recognised by their larger size and dense staining properties. The initial soon divides by a radial wall resulting in two juxtaposed cells. Subsequently, these cells divide repeatedly by



Figs. 1-17. *C. gynandra*. 1-7. Successive stages in the development of campylotropous ovule; 8 L.S. part of integument at megaspore mother cell stage; 9, 10. L.S. part of integument at binucleate and mature embryo sac stages respectively; 12. Cross section of ovule after fertilization. 11, 13. L.S. part of developing seed coats; 14, 16. L.S. ovules at the globular and heart-shaped embryo stages respectively; 15. 17. A portion of seed coat enlarged from 14 and 16 respectively. (ar – archesporium; ce – cellular endosperm es – embryo sac; f – funicle; h – hypostase; ii – inner integument; iii – initials of inner integument; ioi – initials of outer integument; mmc – megaspore mother cell; n – nucellus; ne – nuelear endosperm; oi – outer integument; t – tannin; v – vasculature)

the transverse walls culminating in the production of two layered integuments (Figs. 4, 5). Thus initially both the integuments except the ventral half of outer, consist of two layers of cells. Growth of the integuments is rapid on the dorsal side compared to that on the ventral side resulting in the curvature of the ovule. Although the outer integument initiates slightly later than the inner, it overgrows and contributes to the exostome (Fig. 5). The inner integument forms the endostome only after the exostome is completely organised. The micropyle in all the species is zig zag except in *C. viscosa* where it is straight. Due to repeated mitotic activity the apices of both the integuments become massive (Figs. 6, 7).

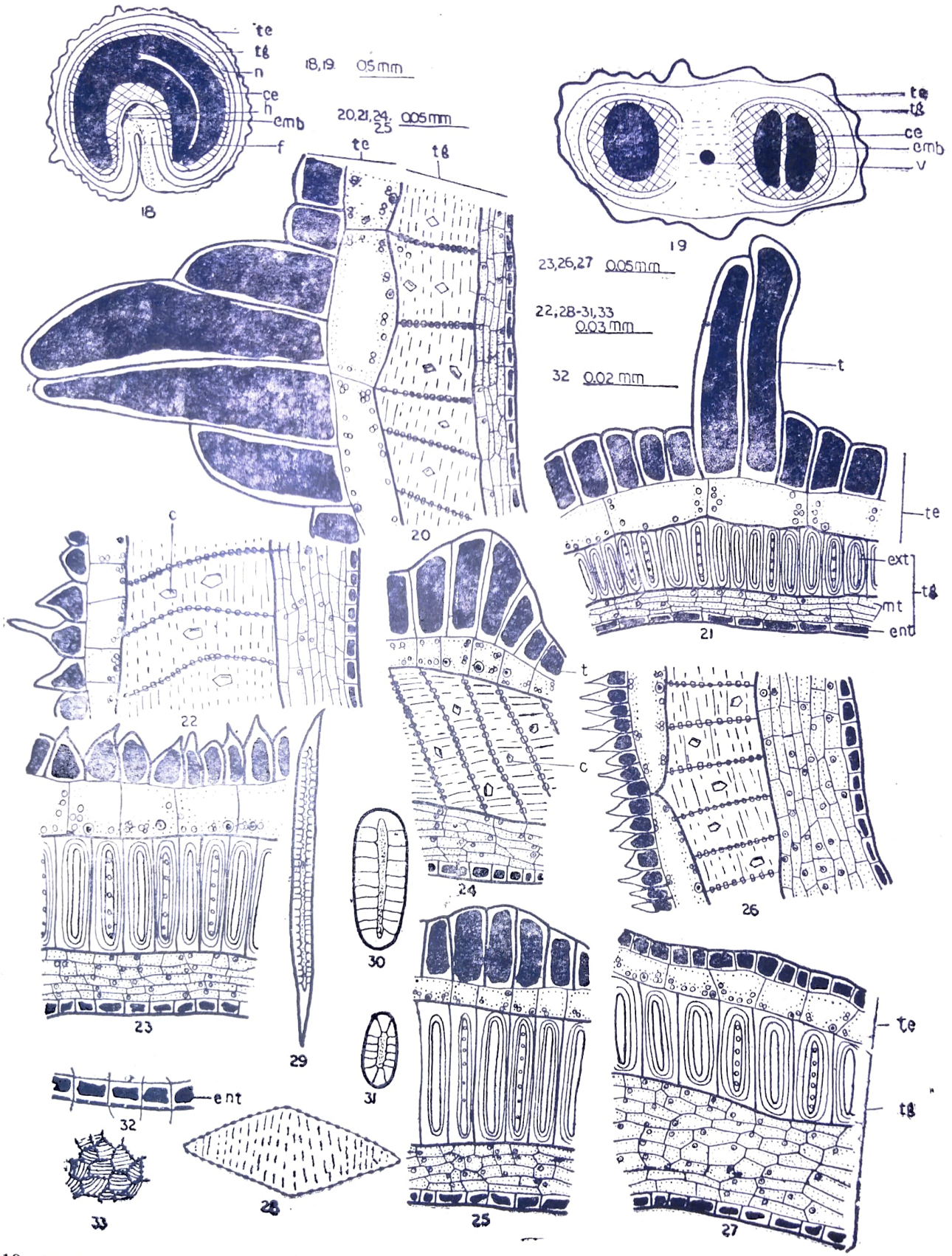
The outer integument remains two layered throughout except at the tip where it is bulbous. The inner integument becomes three layered at the binucleate embryo sac stage and this is attained by the division of the inner layer of the integument (Fig. 9). Simultaneously with the organisation of mature embryo sac, the middle layer divides periclinally to form a four layered inner integument (Fig. 10).

The primary parietal cell that is cut off from the archesporial cell divides to form a parietal tissue of 2-3 layers thick. Divisions in the parietal cells and the cells of the young nucellus result in the formation of a massive nucellus (Fig. 7).

The curvature of the ovule becomes discernible after the initiation of the integuments. This curvature is further accentuated due to differential growth of the integuments which is more pronounced on the convex side. This unilateral growth results in an anatropous condition by the time a four nucleate embryo sac is formed (Fig. 6). This change is supplemented by asymmetrical growth of the nucellus. Owing to this differential growth, the ovule becomes campylotropous (Fig. 7). A single vascular trace is supplied to each ovule and it ends near the chalaza.

Development of seed and seed coat The curvature of the ovule is further accentuated after fertilization by the rapid growth of cells at the funicle and chalaza and this region subsequently forms the raphe. Following fertilization mitotic activity is initiated in the nucellus both at the micropylar and chalazal ends. However it is extensive at the chalazal region and the nucellus becomes curved. The nucellus is absorbed by the growing embryo sac and 1-3 layers of it persists as perisperm in the mature seed. A hypostase is differentiated above the ovular vascular strand (Figs. 14, 16). The inner integument which is referred to as tegmen is multiplicative and becomes 6-9 layered due to mitotic divisions in the cells of the middle layers (Figs. 11-13). Cell differentiation commences and at about the globular embryo stage, the tegmen is differentiated into 3 zones. The cells of the outer epidermis constitute the exotegmen, while those of the inner epidermis constitute the endotegmen. The middle 4 to 7 layers are referred to as the mesotegmen.

Exotegmen The cells of exotegmen are elongate with tapering ends which, when mature are without a protoplast. The walls are hard and lignified. The ends of the exotegmic cells do not taper uniformly to the tip in all planes, but the tapering is confined to the radial plane only. (Fig. 29). The tapering is seen in tangential sections of the cells; transverse sections do not show tapering. The ends of the cells in such sections being rectangular give the appearance of a palisade (Figs. 21, 23, 25, 27). Cell walls exhibit diverse forms of thickenings. The thickenings may be in the form of fine pores as if it were heavily reticulate or they may be scalariform (Figs. 20, 22, 24, 26). The lumen of the cells is large. At the chalazal and micropylar regions the exotegmen is made of small sclerotic cells (Figs. 30, 31).



Figs. 18–21, 28, 29 – *C. gynandra;* Figs. 22,23,30,31 – *C. aspera;* Figs. 24,25 – *C. chelidonii;* Figs. 26,32,33 – *C. monophylla;* Fig. 27. *C. viscosa.* 18,19. L.S. and T.S. of mature seeds respectively; 20,22,24, 26. L.S. of mature seed coats respectively; 21,23,25,27. T.S. of mature seed coats respectively; Radial and front view of exotegmic fibre respectively; 30,31. Sclereids; 32. L.S. endoteg mic layer; 33. Endotegmic cells in surface view. (c–crystals; ce–cellular endosperm; emb–embryo; ent–endotegmen; ext–exotegmen; f–funicle; h–hypostase; mt–mesotegmen; n–nucellus; sg–starch grains; t–tannin; te–testa; tg–tegmen; v–vasculature)

Endotegmen The cells of the endotegmen show slight tangential elongation and develop tannin. The walls of these cells become lignified and their inner tangential walls show minute pits (Fig. 32). These cells are clearly represented in surface view (Fig. 33).

Mesotegmen The cells of the 5 to 8 middle layers remain thin walled. These cells elongate and become vacuolated at maturity (Figs. 20-27).

Testa The outer integument referred to as testa after fertilization is non-multiplicative and remains two layered up to maturity. The outer epidermal cells vary in size and shape in all the species. They appear as an uneven palisade of columnar cells in *C. chelidonii* and *C. gynandra* (Figs. 20, 21, 24, 25). The epidermal cells in *C. aspera* and *C. monophylla* are papillose (Figs. 22, 23, 26) while those of *C. viscosa* are cuboidal (Fig. 27). Subsequent to fertilization, the epidermal cells become thick walled. At about the heart shaped stage, the epidermal cells of the testa develop tannin which is a characteristic feature of the family (Figs. 17, 20-27). The testa in a mature seed becomes ridged due to localised growth of the tegmic fibres (Figs. 18, 19).

The inner epidermal cells of the testa are hyaline, thin walled and are filled with starch. These cells are very much elongated in *C. viscosa*, *C. gynandra* and *C. monophylla* (Figs. 20, 26) and remain small in *C. aspera* and *C. chelidonii* (Figs. 22–25).

The mature seed is reniform and possesses 1-3 layers of perisperm and 2-4 layers of endosperm. The embryo is curved.

DISCUSSION From the observations it is evident that the trizonate ovular primordium and dermal initiation of the integuments are characteristic to the family Capparaceae.

The tegmen in all the members of the family is differentiated into 3 zones. The outer fibrous layer constitutes the exotegmen, the middle thin walled unlignified cells constitute the mesotegmen and the inner delimiting layer with lignified cells constitutes the endotegmen.

Guignard (1983), Orr (1921a) and Raghavan (1937) described the exotegmen as consisting of sclereids. Corner (1976) on the other hand pointed out that the exotegmen in *Cleome* is single layered with laterally compressed ribbon shaped fibres. A close study of microtome sections and seed macerations in the *Cleome* species revealed that the exotegmic cells differ from the fibres on 2 grouuds (1) the presence of a large lumen and abundant pits, (2) the end of cells does not taper uniformly to the tip in all planes.

The cells of the exotegmen retain their nuclei till a late stage and thay are largely vacuolate and on this account Orr (1921a) described this layer as the aqueous tissue. Raghavan (1937) considered this layer as a protecting envelope, preventing dessication of the embryo. The presence of a large lumen and abundant pits suggests that the exotegmic cells are structurally adapted, to the function of conduction.

The cells of the endotegmen differ from the rest of the cell layers by their lignified cell walls and presence of tannin. Baillon (1872) and Orr (1921b, c) stated that the ovules and seeds of Capparaceae are unique in possessing a highly specialised tissue which is adpressed to the inner wall of the integument on one side and is in close eontact with the embryo on the other side. They have described this layer as a third seed coat. Since the cells of this layer possess the form and structure of tracheids this specialised layer is termed by Orr (1921c) as the trachael sheath. Orr (1921b, c) believed that this tissue actively functions as an accessory water supply system for the embryo and forms a kind of water jacket surrounding the embryo.

Arunalakshmi (1985) after studying the development of integuments in 7 species of the family con-

cluded that there is no so-called third seed coat in the family as reported by Baillon (1872) and Orr (1921b, c). Her studies confirmed the opinion of Raghavan (1937) who regarded the so-called third integument as nothing but the enlarged cells of the inner epidermis of the tegmen whose walls had become lignified. Since the observations of Orr (1921b, c) are confirmed to the peels of mature seeds and no sections have been taken it is probable that a wrong impression was gained by Baillon (1982) and Orr (1921b, c) that these cells represent an additional seed coat.

The seed coat development is similar in all the species and from the hitherto available information it is evident that mode of development of seed coat is useful in classification only at generic level (Arunalakshmi, 1985). The architecture of the mature seed coat is however useful to some extent for identifying species. The epidermal cells of testa are columnar in *C. gynandra* and *C. chelidonii* while those of *C. monophylla* and *C. aspera* are papillose. In *C. viscosa* the epidermal cells of testa are cuboidal.

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