CYTO-MORPHOLOGICAL FEATURES OF *PORTULACA TUBEROSA* ROXB.

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1. Introduction

THE family Portulacaceæ has been studied to some extent by previous investigators. The earliest worker in this family is according to Schnarf (1931), Hofmeister (1858) who studied the embryology of *Calandrinia*. Later d'Hubert (1896) investigated two other genera, *viz.*, *Portulaca* and *Talinum*, in addition to *Calandrinia*. Rócen (1927) made a comparative study of the embryology and development of female gametophyte in about eight genera, belonging to this family including *Calandrinia*, *Claytonia*, *Montia*, *Portulaca*, and *Anacampseros*. In all these, he found the normal type of development.

Suguira (1936) gives a brief account of the previously determined chromosome numbers in this family. He has also reported the haploid numbers of *Calandrinia speciosa* and *C. umbellata* as twelve and ten respectively.

The genus *Portulaca* has also received some attention. Rócen (1927) studied the development of embryo-sac and embryo in *Portulaca grandiflora.* Later Cooper (1935 and 1940) studied the microsporogenesis and embryology of *Portulaca oleracea* whose haploid number, he reported as 27.

In this paper, the cytological and morphological features of *Portulaca tuberosa* Roxb., are described. Chromosome numbers of some of the local species of *Portulaca* are also given, having been determined for the first time.

2. *Materials and Methods*

Portulaca tuberosa Roxb., is a common perennial weed, with short stems spreading from a tuberous root. The root is 2–3 inches long and is slightly fusiform with a few branches towards the extremity. The leaves are arranged alternately on the spreading stem which is 2–3 inches long. The leaves are linear and fleshy with nodal appendages of sparingly tufted brown or silvery hairs. Flowers are in small clusters, surrounded by 7–10 leaves and tufted hairs and are yellow in colour. Stamens are many and the style is filiform and five-cleft.

Root-tips were collected from plants grown in pots and fixed in Müntzing's modification of Navaschin's fluid (Müntzing, 1933). For meiotic stages, whole buds were fixed with or without prefixation in Carnoy's fluid. Stages for morphological studies were obtained from small buds and ovaries fixed in formalin-acetic alcohol. The materials were embedded in paraffin wax using chloroform as the paraffin-solvent. Sections were taken at thicknesses varying from 6–15 microns and were stained in iron-alum hæmotoxylin and Newton's iodine Gentain-violet.

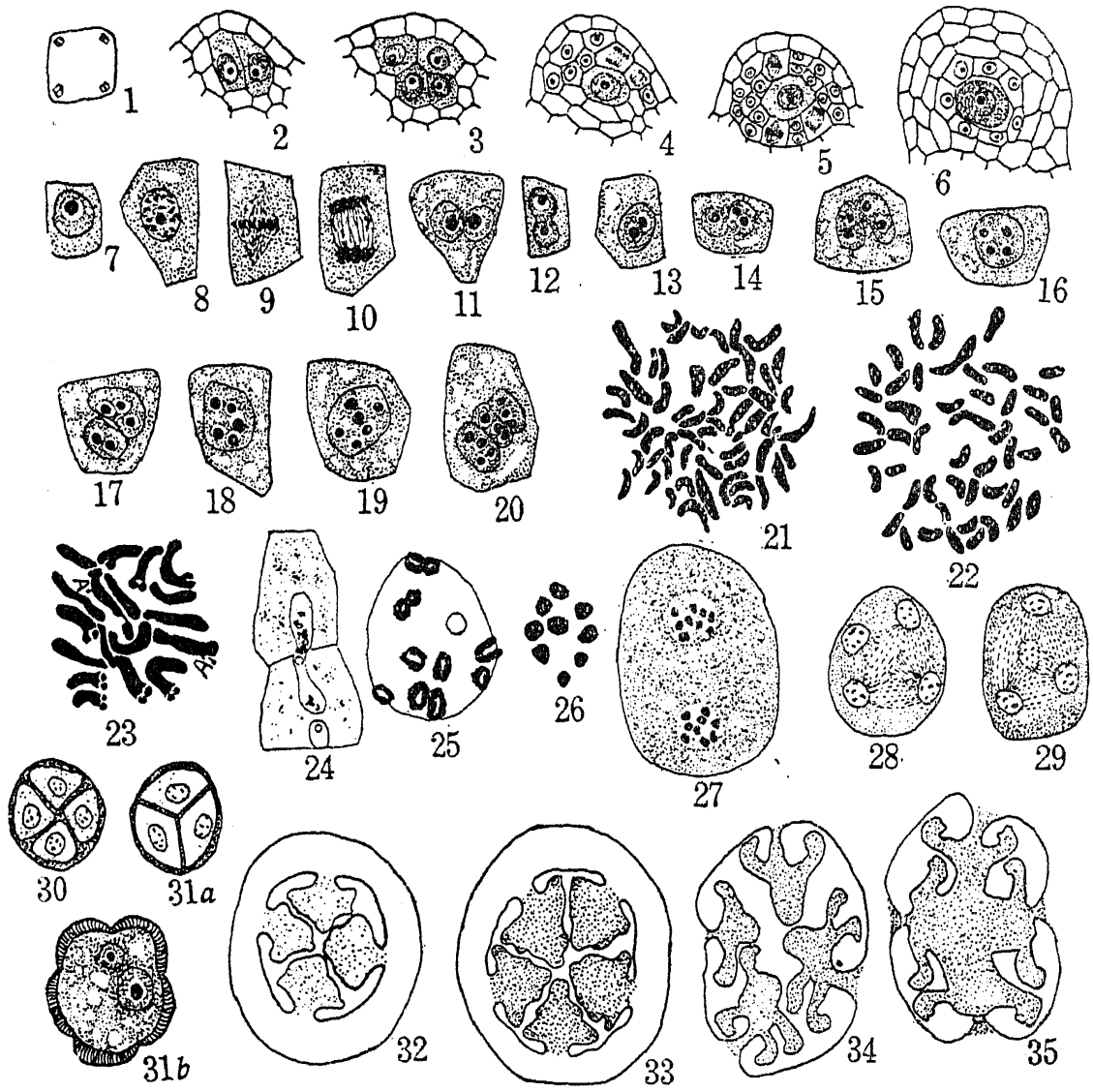
3. *Observations*

(*a*) *The microsporangium :*

The archesporium of the anther is differentiated at the four corners of the anther, when the four lobes have not been formed (Fig. 1). The archesporium consists of two cells which are hypodermal in position (Fig. 2). Cooper (1935) reports that a band of seven to eight cells constitute the archesporium in *Portulaca oleracea.* Archesporial bands such as are met with in the present case, have been observed in the genera *Bergia, Sesuvium* (Raghavan and Srinivasan, V. K. , 1940, *a* and *b*) and in *Spermacoce* (Raghavan and Srinivasan, A. R., 1941, *c*).

Through periclinal divisions of the archesporial cells, two layers are formed, the outer being the primary wall layer and the inner the primary sporogenous layer (Fig. 3). By further divisions of the primary wall layer, more parietal layers are formed surrounding the sporogenous cells (Figs. 4–5).

The sporogenous cells do not divide much and hence only one pollen mother cell is seen in each lobe of the anther in transverse sections. Presumably there is only one row of microsporogenous cells. We find this confirmed in longitudinal sections. At the mature stage, the wall of the anther consists of four layers, the innermost of which functions as the tapetum (Fig. 6).



TEXT-FIGS. 1–35. *Portulaca tuberosa*

FIGS. 1–35 represent stages of microsporogenesis and development of placenta. FIG. 1. Transverse sections of young anther. × 75. FIG. 2. Archesporium of the anther. × 750. FIG. 3. The primary wall layer and the primary sporogenous layer. × 750. FIGS. 4 and 5. Division of cells in wall layers forming more parietal layers. × 750. FIG. 6. Anther showing three wall layers and the tapetum. × 750. FIG. 7. Tapetal cell with a single nucleus. × 1500. FIGS. 8–10. Stages of division (mitotic) of the tapetal nuclei. × 1500. FIGS. 11–20. Various stages of fusion of tapetal nuclei. × 1500 (explanation in the text). FIG. 21. Somatic metaphase plate of *Portulaca* sp. × 3900. FIG. 22. Diploid chromosome complement of *Portulaca quadrifida*. × 3900. FIG. 23. Somatic plate of *Portulaca tuberosa*. × 3900. FIG. 24. Cytomixis in *P. tuberosa*. × 1100. FIG. 25. Diakinesis. × 3900. FIG. 26. Meiotic M I plate. × 3900. FIG. 27. M II plate. × 3900. FIGS. 28 and 29. Spindle formation in T II. × 1100. FIGS. 30 and 31 *a*. Isobilateral and tetrahedral tetrads. × 750. FIG. 31 *b*. Mature pollen grain. × 750. FIGS. 32 and 33. Early stages of placenta formation. × 150. FIG. 35. Placentas growing towards the centre. × 75. FIG. 35. Showing the central tissue formed by the fusion of the placentas. × 75.

The behaviour of the tapetal nuclei is worth mentioning. To begin with, the tapetal cells are uninucleate and are full of cytoplasm (Fig. 7). Later, small vacuoles arise in the cytoplasm and the nucleus shows signs of division (Fig. 8). The division of tapetal nucleus takes place even before the nuclei of the pollen mother cells enter into the early meiotic stages. Cooper (1935) observed that the division took place while the nuclei of the microspore mother cells were passing from synizesis into the heterotypic plate. The division in this case is mitotic as can be seen from Figs. 9–10. Some workers have reported amitosis as the typical method by which the tapetal nuclei divide. Such a case has been reported by O'Neil (1920) in *Datura stramonium*. Rócen (1927) also reports a similar amitotic division in *Portulaca grandiflora*. Cooper (1935), however, found that the tapetal nuclei in *Portulaca oleracea* multiply through ordinary mitotic division. These and other recent observations in many other genera distributed over a wide range of families can be taken as additional evidence to support the view that the nuclei of the tapetal cells divide mitotically. Cooper (1933) recognises three types of tapetal cells: (1) in which the tapetal cells remain uni-nucleate, (2) in which they are bi-nucleate and (3) where they become pluri-nucleate. The present case belongs to the third type and shows the interesting phenomenon of tapetal nuclear fusion. It is possible to trace through intermediary stages how multi-nucleolate nuclei arise. Fig. 11 shows a bi-nucleate tapetal cell and Fig. 12 represents the fusion of the nuclei. In Fig. 13, a tapetal cell with a two-nucleolated nucleus is shown. This is the result of the above said fusion. Fig. 14 represents the fusion of a bi-nucleolated nucleus with an ordinary nucleus to result in a three-nucleolated nucleus (Fig. 17). Two tapetal nuclei each having two nucleoli by fusing give rise to a four-nucleolated fusion nucleus (Figs. 15 & 16). The fusion of three-nucleolated nucleus and a two-nucleolated nucleus is shown in Fig. 17, which results in the formation of a five-nucleolated nucleus (Fig. 18). A peculiar case was found in which three nuclei were seen to fuse (Fig. 20). Of the three nuclei, two had three nucleoli and the remaining one, four nucleoli. Such a fusion will result in a nucleus having ten nucleoli. A completely fused ten-nucleolated body was, however, not met with. But as in Fig. 19, six-nucleolated fusion nucleus was not uncommon.

Fusion of tapetal nuclei resulting in the formation of multi-nucleolated nuclei have been met with in many cases. Such a phenomenon has been recorded in *Nicotiana glutinosa* (Raghavan and Srinivasan, A.R., 1941 *b*), in *Ranunculus* (Singh, 1936) and *Chenopodium* (Bhargava, 1936). It is interesting to note that Cooper (1935) found that in *Portulaca oleracea*, the tapetal cells remained bi-nucleate till their final degeneration. We are not told whether these fuse at all.

The pollen mother-cells round off and show starch grains in their cytoplasm. Such accumulation of starch grain in P.M.C.'s has been recorded in *Spathodea* (Raghavan and Venkatasubban, 1940).

(*b*) *Somatic chromosomes*:

The diploid chromosome number of this species is shown in the somatic plate in Fig. 23. There are 18 chromosomes of which one is found to have a satellite at its distal end connected by a trabant. Perhaps the body is too big to be called a satellite. It may be more appropriate to describe the chromosome as having a secondary constriction, the primary constriction being subterminal. Very likely the homologue of this chromosome (A) is A_1 where however the trabant is not so prominently seen owing to a possible twist and the orientation of the chromosome. Two of the chromosomes are comparatively short.

(*c*) *Meiosis*:

During early prophase stages the extrusion of chromatin material from one cell to its neighbouring pollen mother-cell, was observed in some cases. Such extrusion is commonly known as cytomixis. In this case, the prophase threads formed a sort of wad and this chromatin lump was extruded along with the nucleolus into the adjacent cell (Fig. 24). Such a phenomenon has been met with in the interspecific hybrid between *Nicotiana tabacum* × *N. glutinosa* and its importance has already been discussed (Raghavan and Srinivasan, A. R., 1941 *a*).

Cytomixis generally is the cause of meiotic abnormalities, obviously due to a disturbance of the chromosome balance in the pollen mother-cells. Though the phenomenon occurred here and there, not many abnormalities worth recounting were met with, so far as the meiotic process is concerned.

During diakinesis nine bivalents are formed (Fig. 25). The bivalents are fairly large. Two of the bivalents possess three chiasmata, two terminal and one interstitial. Six ordinary ring bivalents are met with, each with two terminal chiasmata. Only one bivalent showed a single terminal chiasma, which is obviously the result of the pairing of the two shortest chromosomes. The nuclear membrane disappears and the nucleus after a short prometaphase enters upon the metaphase. During metaphase, nine bivalents are observed, all arranged in the equatorial plate (Fig. 26). Disjunction seems to be normal and at second metaphase nine chromosomes are seen at each of the poles (Fig. 27).

During second telophase (Figs. 28 & 29) four groups of chromosomes are organised and six spindles are formed connecting each of the telophase

groups with the other three. When all the four groups are arranged in one and the same plane (Fig. 28), there is an isobilateral arrangement which after the process of furrowing gives rise to an isobilateral tetrad (Fig. 30). If however, the telophase groups are situated at right angles to one another, the tetrahedral type is the result (Figs. 29 & 31*a*). The furrowing takes place simultaneously. The furrows formed are very narrow during initial stages and the process is apt to be construed as cell-plate formation. This however is not the case in the present investigation, for, not only has actual furrowing been observed, but other structural details necessary for cell-plate formation are not present also. For instance, if organisation of the tetrads was by cell-plate formation, then a definite thickened portion formed by the aggregation of small particles of cell wall matter must be observable in the middle of each spindle. The spindle itself should have been greatly stretched laterally. These were not observed in the present case.

Healthy pollen grains of large size are formed which are two celled at the shedding time (Fig. 31 *b*). Cooper (1935) reports that the pollen grains are three-nucleate at the shedding time.

(*d*) *Development of the placenta and the ovules :*

The ovary is unilocular and half inferior. It is made up of two to five carpels. A somewhat critical ontogenetic study of the ovary reveals some interesting facts. In the earliest recognisable stage the gynæceum, formed by the marginal fusion of the four carpels (Figs. 32 & 33), shows parietal placentation. The primordia of the ovules look like small knob-like protuberances. At a later stage the tips of these carpels namely, the non-ovule bearing conical tips of the placenta, come together and fuse; so much so, a structure that may be regarded as axile placenta is formed (Figs. 34 & 35). After this the placentas gradually lose their connection with the ovary wall so that the ovule seems to arise from placental structures which are free from the surrounding wall of the ovary. Strictly speaking the wall of the ovary is composed of the four or five carpels which are fused together to form a tubular structure. Internal to this there is this placental region bearing the ovules. This is a condition which simulates a free-central placentation but which cannot be strictly regarded as such, as it would mean an ovule bearing tissue taking its origin from the base of the ovary. The points of interest that may be raised in passing are (1) whether this is truly a case of free-central placentation, and (2) if that is so can the developmental history of the placenta throw any light upon its derivation. If the recapitulation theory is applicable in this case, then we can say that the order of development is parietal, axile and free central. A generalisation on this matter must however await

a more critical and widespread ontogenetic study of the members especially of the Centrospermeæ.

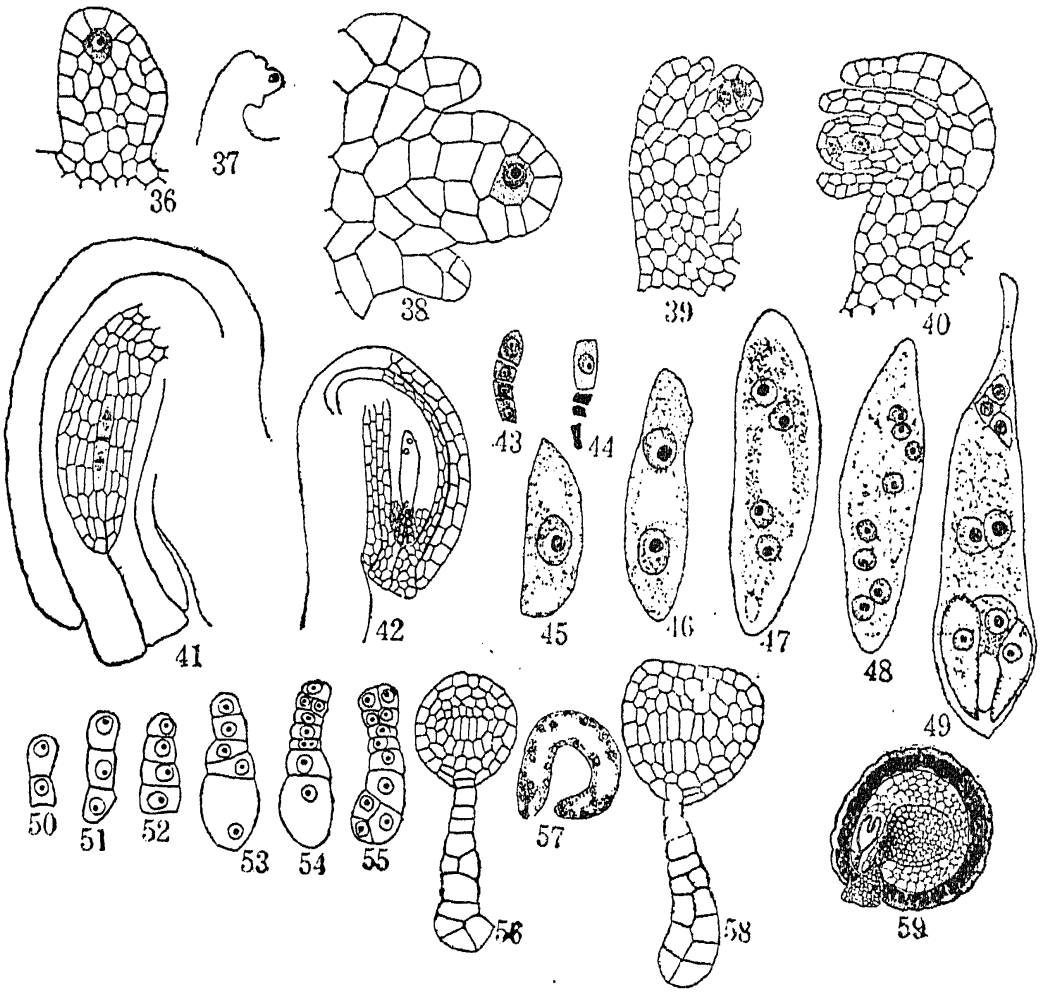
The archesporium of the ovule is differentiated when the ovule arises as a straight protuberance from the placenta (Fig. 36). It consists of a single hypodermal cell larger than the surrounding cells rich in cytoplasm and having a prominent nucleus. During the growth of the ovules, the funicular portion elongates and the body of the ovule bends towards the one side (Fig. 37). Later the archesporium divides periclinally to result in the primary parietal cell lying on the outside and the primary sporogenous cell inside (Fig. 39). The primary sporogenous cell without further division elongates and functions as the megaspore mother-cell and is invested by two layers of nucellus on its side, capped by the primary parietal cell at the top (Fig. 40). The primary parietal cell gives rise to three or four layers of wall cells so that at later stages the dyad and the embryo-sac come to be situated three or four layers deep in the nucellus (Figs. 41 & 42). The nucellar cells at the sides of the megaspore mother-cell also divide causing the ovules to become massive. During the early embryo-sac stages the ovule is anatropus (Fig. 42). But later it bends on itself to give rise to the curved embryo-sac and thus forms a campylotropous structure. In the mature ovule (Fig. 59) the ovule becomes so curved that the micropylar and the chalazal ends nearly meet and most of the funicular tissue is caught in between them.

The two integuments make their appearance when the ovule begins to curve towards one side (Fig. 37). The inner integument is the first to make its appearance (Fig. 38). It is at first two layers thick (Figs. 38–40). It continues to remain so during later stages also, except at the micropylar region where it is three or four layers thick (Fig. 42). The inner integumental halves meet at the micropylar region and constitute the micropyle. The outer integument is differentiated soon after the inner integument. But that half of the outer integument on the side of the ovule towards the funicle is not differentiated until late in the development of the megaspore mother-cell (Figs. 39–41).

The outer integument even at later stages does not overtake the inner integument and therefore cannot be said to contribute to the organisation of the micropyle. It remains two layered throughout.

(*e*) *Megasporogenesis :*

The nucleus of the megaspore mother-cell undergoes meiosis and results in the dyad. The cells of the dyad are more or less of equal size (Fig. 41). Cooper (1940) however has found in *Portulaca oleracea* that the chalazal cell is larger and increases in size during interkinesis. He has also reported that the spindle in the micropylar cell may be at any angle. In this species,



TEXT.-FIGS. 36–59. *Portulaca tuberosa*

FIGS. 36–59 show the development of embryo-sac and embryo. FIG. 36. Archesporium of the ovule. × 750. FIG. 37. The ovule at a later stage. × 75. FIG. 38. The same magnified showing the development of integuments. × 1500. FIG. 39. Primary parietal and primary sporogenous cells. × 750. FIG. 40. Megaspore mother-cell. × 750. FIG. 41. Ovule at the dyad stage. × 750. FIG. 42. Ovules at the 4-nucleate embryo-sac stages. × 350. FIGS. 43 and 44. Linear tetrads. × 750. FIGS. 45–47. Embryo-sac stages. Explanation in the text. × 1500. FIG. 48. Eight-nucleate embryo-sac at an early stage. × 1100. FIG. 49. Mature embryo-sac. × 1500. FIGS. 50–56. Embryo stages. × 750. FIG. 57. Shows the nuclear endosperm. × 75. FIG. 58. Embryo just before lobing of cotyledons. × 750. FIG. 59. Section of a young seed. × 75.

however, such is not the case. Even during homotypic division the dyad cells are of equal size and both the spindles are in a straight line (Fig. 41). Cooper (1940) adds that the division in the micropylar cell lags behind that of the chalazal cell. In the present case we find simultaneous division resulting in a linear tetrad (Fig. 43) of megaspores. The chalazal megaspore develops while the other three degenerate (Fig. 44). The megaspore develops into the uni-nucleate embryo-sac (Fig. 45). The nucleus divides and the daughter nuclei travel to the opposite poles and the two-nucleate embryo-sac is formed (Fig. 46). Another division of the embryo-sac nuclei results in the formation of the four-nucleate embryo-sac (Fig. 47). Starch grains are formed at this stage in the embryo-sac and the cytoplasm is very vacuolate. The four

nuclei divide to give rise to eight-nucleate embryo-sac. Of the eight nuclei, one from each pole travels towards the centre to fuse later into the secondary nucleus (Fig. 49). Thus there are three nuclei left at each of the poles. The three nuclei at the micropylar end are bigger than those at the chalazal end.

The mature embryo-sac is long and tapering at the antipodal end (Fig. 49). It consists of the egg-apparatus at the micropylar end, the antipodals at the chalazal end and the polar nuclei in the centre. The egg-apparatus consists of the synergid cells, the ends of which lie apart from each other during early stages but come closer during later stages. The synergids are large, each with a basal vacuole and the nucleus above the vacuole. No starch grains were found in the synergids. Cooper (1940) has found a distinct filiform apparatus in the synergids of *Portulaca oleracea.* The egg nucleus is much larger than the nuclei either of the synergids or of the antipodals. The egg cell has a large apical vacuole and its cytoplasm contains inclusions of starch grains. The antipodal cells are three in number and are small in size. They are arranged in the form of the letter V. In *Portulaca oleracea* they were found to be located in an axial row at the chalazal end. These are ephemeral and degenerate before fertilization.

(f) Endosperm and embryo :

After fertilization, the endosperm nucleus divides and gives rise to free nuclear endosperm. The endosperm remains nuclear until a spherical mass of embryo cell is formed (Fig. 57). Cell-wall formation takes place only later resulting in loose cells.

The oospore undergoes a transverse division followed by cell-wall formation. Thus a two-celled pro-embryo is formed (Fig. 50). Through further divisions the three, four and five-celled pro-embryos are formed (Figs. 51–53). The basal cell and the cell next to that increase in size (Figs. 53 & 54). After a linear pro-embryo of seven cells is formed, the cell next to the apical cell undergoes an anticlinal division giving rise to the quadrant (Fig. 54). Thus the embryo at this stage consists of the quadrant and a uniseriate suspensor five cells long. The cell of the suspensor next to the quadrant becomes the hypophysis, while the quadrant through the anticlinal division of the apical cell, results in the formation of an octant (Fig. 55). At this stage the lowermost cell of the suspensor undergoes a periclinal division resulting in a small basal cell and a large apical cell, the latter of which divides anticlinally. Thus the basal cell of the suspensor becomes biseriate during the octant stages.

Periclinal and anticlinal walls are laid in the octant and a spherical mass of cells is formed (Fig. 56). Anticlinal divisions in the basal suspensor cells

take place so that at a later stage each of the five basal rows of suspensor cells come to be made up of two cells (Fig. 58).

Fig. 59 shows a young seed in which the cotyledonary lobing of the embryo has been effected. The endosperm is cellular and soon the embryo eats up the whole endosperm. The mature seed is thus exalbuminous. The testa consists of two layers of which the inner layer is sclerotic.

(*g*) *Some chromosome numbers in the genus,* Portulaca *:*

Portulaca sp.—54 Chromosomes were counted in somatic metaphase plates (Fig. 21). Many of the chromosomes are short. Two of the medium-sized chromosomes show satellites.

Portulaca quadrifida Linn. (Fig. 22).—Sections of root-tips show metaphase plates with forty-eight chromosomes. All the chromosomes except two are short.

4. Discussion

(*a*) *The behaviour of tapetal nuclei; its possible meaning*:

In a number of genera investigated in this laboratory, it has been found that the division of the nuclei of the tapetum (of the microsporangium) and their ultimate fusion prior to disintegration is almost the rule. The real significance of this repeated division of the tapetal nuclei and their later fusion is not quite clear. That the cells of the tapetum are nutritive in function, there can be no doubt. If this is conceded, we have to interpret the division and fusion of the nuclei only on the basis of this primary nutritive rôle. Possibly, an analogy can be drawn from that important nutritive tissue in angiosperms, the endosperm, where it is the result of repeated division of the triple fusion nucleus. First of all there is a fusion of three nuclei, two polars and the male, and then a subsequent division, either into free-nuclei or a cellular structure, to form the so-called endosperm tissue. It may be that in the tapetal cells, nutritive as they primarily are, a sort of a fusion of nuclei must precede the formation of nutritive materials ; hence the nuclei divide. This not only increases nuclear material, *i.e.*, the nutritive material, but also acts as a stimulus even as in the case of the triple fusion nucleus. The tapetal nuclear behaviour may be regarded in some measure as a simulation of the triple fusion nucleus. Seldom do we find the divided nuclei in a separate condition. In other words, immediately upon their division they fuse. While it is likely as has been mentioned above, that this fusion is a simulation of the sexual act which precedes the formation of the other nutritive tissue, it is likely that it is also governed by spatial considerations. Obviously, the fusion product of four or five nuclei occupies far less space than five separate nuclei, hence perhaps this immediate fusion.

(*b*) *Morphological details; are there any too minor to recount?*

In the course of cyto-morphological investigations upon different plants, there is a wide measure of uniformity so far as broad details of micro- and macro-sporogeneses are concerned. The question is pertinently asked, whether there is any need to recount all these details in different genera though they may belong to different widely separated families. In essence the morphological details are the same; why then should one almost repeat these details in various genera? The answer for this question must be found in another question; are these morphological details (pertaining to the ontogeny of the micro- and mega-gametophytes) of any value taxonomically and phylogenetically? or, are they merely descriptive and devoid of any such significance? To our mind the former seems to be more likely.

It is interesting that what may be regarded as important features are not characteristic even of a genus, not to speak of a family. While in the course of investigation we find some features characterising almost all the members of a particular genus and even members of the family, we find at the same time that, species belonging to the same genus show wide variations in respect of features which at first sight may be regarded as important. So far as *Portulaca* is concerned, the present species *P. tuberosa* differs from the previously investigated *P. oleracea* in some important details. For instance, in *P. oleracea* only bi-nucleate tapetal cells were found, whereas in the present species multinucleate tapetal cells are the rule. In *P. grandiflora* multinucleate tapetal cells were met with. The condition of the microspore in the present species and *P. grandiflora* is two-nucleate at the time of shedding whereas it is three-nucleate in *P. oleracea.* It only shows that so far as these morphological details are concerned there cannot be any type feature that could be said to be characteristic either of the genus or of the family. That could be found out only by a critical examination of a large number of species belonging to the genus or of a number of genera comprising a family. If at the end of such a study we find some features common throughout, then only can it be said that they are characteristic. It might well be that what may at first sight be regarded as insignificant details may be of considerable importance from this point of view. An instance of this kind may be drawn from the critical studies of Aizoaceæ (Raghavan and Srinivasan, V. K., 1940 *b*); it was found that the two tribes of the Aizoaceæ, the Molluginoideæ and the Ficoideæ, were characterized by a number of morphological features, some of which like the presence of the aril and its absence may rightly be considered as important morphological characters. But alongside of this, such minor features as the radial elongation of the epidermal cells of the

nucellus, the number of layers constituting the integument, etc., were also found to be useful in determining the characteristic features of the two sub-groups. Another instance of this kind can be had in the investigations in the family Elatinaceæ (Raghavan and Srinivasan, V. K., 1940 *a*) where, morphological details such as the order of wall formation in the endosperm tissue have been made use of to support the division of Elatinaceæ into two sub-groups, the Elatinoideæ and the Bergioideæ, which division has been made, on the basis of chromosome numbers and other floral characters. Similarly, in the Rubiaceæ (Raghavan and Srinivasan, A. R., 1941 *c*) such apparently insignificant details as the number of cells composing the nucellar epidermis, the configuration of the epidermis, etc., which might well be passed over in a casual study, were upon a critical examination, found to play an important rôle in the matter of the evolution of chromosome numbers in the family. All these only go to show that however minor and insignificant any morphological detail may be from a superficial point of view, it may be of significance for purposes of generalization which, however, means a more critical and wide study of a particular group. In other words no morphological detail is too insignificant to be recorded. On the other hand important characters may have no morphological value in a general critical study either of a genus or of a family.

(*c*) *The position of Portulacaceæ :*

A word may now be said of the position of Portulacaceæ about which there is difference of opinion. Taxonomists have assigned to it one position or another based almost solely upon floral characters and some vegetative features. A few like Puri and Singh (1935) have assigned to it another position mainly upon the basis of anatomical features. No one has so far tackled this problem from the point of view of chromosome numbers. As cyto-taxanomy is coming to play an increasingly important part, it would be well worth our while to review the position of the *Portulacaceæ* from this point of view also and try if possible, to correlate the position indicated by chromosome numbers, with other data on the basis of which the taxonomical position of this family has so far been determined.

Bentham and Hooker (1862–1883) have grouped *Portulacaceæ* under *Thalamifloreæ* along with *Caryophyllaceæ* (the other minor families like *Frankeniaceæ*, etc., are not mentioned as they are not necessary for our purpose). Engler and Prantl (1894) placed this family in the *Monochlamydeæ* under the group *Centrospermeæ* along with seven other families showing curved embryos. Hutchinson (1926) however divides the *Centrospermeæ* into two groups, the *Chenopodiales* and the *Caryophyllales*. *Portulacaceæ*,

Caryophyllaceæ and *Aizoaceæ* come under *Caryophyllales* while the rest are grouped under *Chenopodiales.*

Puri and Singh (1935) are of opinion that the families, *Chenopodiaceæ, Amaranthaceæ, Nyctaginaceæ, Phytolacaceæ* and *Aizoaceæ* form a very natural grouping. That is, they separate *Aizoaceæ* from *Portulacaceæ.* It differs from Hutchinson's classification in this important feature. In effect their classification is very like that of Bentham and Hooker (1862–1883), though different names are given to the two groups (*Centrospermales* = *Apetalæ* of Hooker and *Caryophyllales* = the *Polypetalæ* of Hooker). Their opinion is based on some embryological details for which phylogenetic importance has been assigned. They also consider anomalous secondary thickening a very important anatomical character, which being present in *Aizoaceæ,* is the reason for its removal from the *Caryophyllales* of Hutchinson to the Centrospermales where it will fit in better with the other orders showing anomalous secondary thickening. The different systems of classification pertaining to this are roughly summarisd in the accompanying table. Chromosome numbers in the different families are also indicated.

On closer scrutiny it seems that the affinities of *Portulacaceæ* have not been given due consideration, when forming the latter said classification. The external features of *Portulacaceæ* bear a great resemblance to those of *Aizoaceæ.* In habit, both are essentially herbaceous. Both include genera with succulent vegetative parts, circumcessile capsules and above all, curved embryo. The embryological features that these authors report to be characteristic of the families in the *Centrospermales,* are found to occur in *Portulacaceæ* also, with the exception of one or two, *e.g.,* the absence of anomalous secondary thickening, the non-occurrence of periclinal divisions in the nucellar epidermis, and the two-nucleate conditon of pollen in *Portulacaceæ.*

Table representing the position of Portulacaceæ according to various Systems of Classifications

[Haploid chromosome numbers occurring in the families are quoted from Tischler's list of chromosome numbers (Tischler, 1938)]

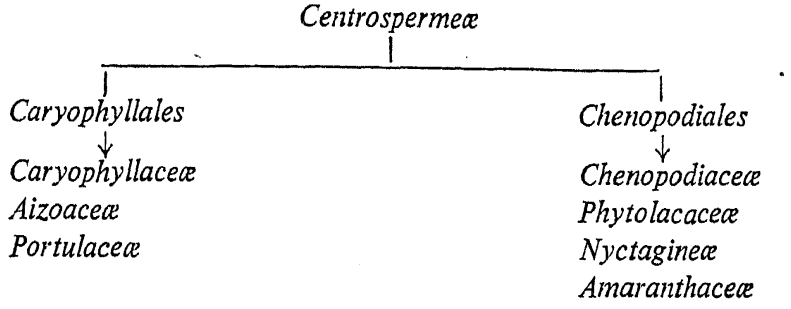
(1) According to Bentham and Hooker (1862–83):

Thalamifloreæ——*Caryophyllineæ*——*Caryophyllaceæ*, n = 8, 9, 10, 11, 12, 13, 15, 30 & 45.
Portulacaceæ n = 8, 9, 10, 18, 24 & 27.

(2) According to Engler and Prantl (1894). Rendle (1938) has in the main followed this classification:

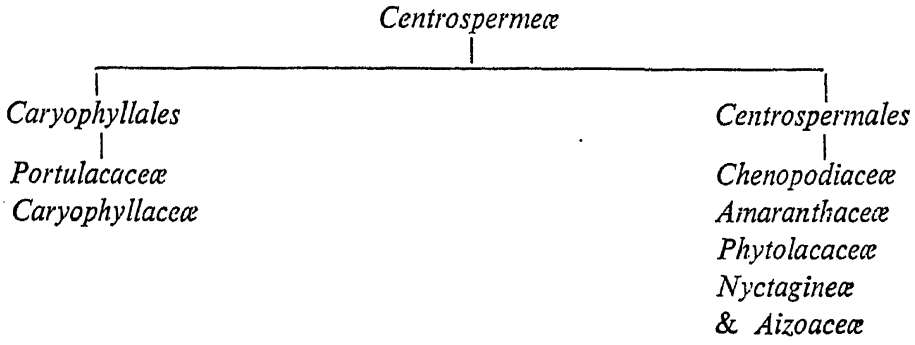
Monochlamydeæ
|
Centrospermeæ
|
Chenopodiaceæ, n = 9 & 18; *Amaranthaceæ*, n = 6 & 21; *Nyctagineæ*, n = 29; *Phytolaccaceæ*, n = 9, 18, 36 & 54; *Aizoaceæ*, n=9, 16 & 32; *Portulacaceæ* (chr. nos. given above); *Caryophyllaceæ* (chr. nos. above).

(3) According to Hutchinson (1926):



(chr. nos. above)

(4) According to Puri and Singh (1935):



Examined in the light of chromosome numbers, two features are found to be most striking, first, the relationship betweem *Portulacaceæ* and the *Centrospermales* of Puri and Singh, and secondly the relative lack of similarity of the chromosome numbers of *Portulacaceæ* and those of the *Caryophyllaceæ*. Considering the first-mentioned feature, we find that the chromosome numbers of *Portulacaceæ* mainly belong to the three and four series. Suguira (1936) after considering the chromosome numbers in the genus *Portulaca* has come to the conclusion that nine is the basic number of the genus. The same author in a later work (Suguira, 1940) observes, "According to Pax, *Portulacaceæ* and *Aizoaceæ* are closely related. Karyologically this can be recognized too, for they both have a theoretical basic number of three." Perhaps from this primary basic number 3, a secondary basic number 4 also arose, of which we get multiples as representatives. Examining the chromosome numbers of the *Centrospermales* in this light, we find that the chromosome numbers of *Aizoaceæ* are multiples of 3 and 4, those of *Phytolaccaceæ*, *Amaranthaceæ* and *Chenopodiaceæ*, multiples of 3 only. *Nyctagineæ* alone shows 29 as the chromosome number of *Mirabilis*. Thus from the point of view of chromosome numbers *Portulacaceæ* seems to be closely related to the members of the *Centrospermales* of Puri and Singh (1935).

Secondly, the grouping of *Portulacaceæ* along with *Caryophyllaceæ* may be considered. Viewed in the light of chromosome numbers, this grouping seems to be unnatural. *Caryophyllaceæ* has been pointed out to be a large and heterogenous family. Puri and Singh (1935) have said that the

largeness and the variety in its structure warrant the elevation of the family to an independent order. This is supported by the nature of the chromosome numbers also. There is a gradation of chromosome numbers from 8 to 13. The majority of the numbers occurring in the polyploid series are multiples of 5 and not of 3 or 4. Thus from the point of view of chromosome numbers the *Caryophyllaceæ* is less related to *Portulacaceæ*, than the families included in the *Centrospermales* are to the latter.

It thus seems to us, that there is not much justification for separating *Portulacaceæ* and *Aizoaceæ*. They conform to one another, not only in chromosome numbers but also in other important details. It would therefore be more in accordance with the data available so far, including chromosome numbers, to remove *Portulacaceæ* from the *Caryophyllales* of Puri and Singh and bring it under the other group along with *Aizoaceæ*. This would mean that *Caryophyllaceæ* would form a group by itself and the rest of the families would constitute the other group. Some data which Puri and Singh (1935) think important for the separation of *Aizoaceæ* from the *Portulacaceæ* are now disclosed by the present investigation to be of no great morphological value. For example, *Aizoaceæ* has three-celled pollen grain while *Portulacaceæ* has only two-celled pollen, and hence it is contended that they cannot be put together. But it has been shown in the present investigation that this is a feature which varies among the members of even a single genus and therefore this cannot be given much importance.

The other alternative is to retain Hutchinson's classification, which would statisfy the important condition of keeping *Portulacaceæ* and *Aizoaceæ* together. But along with these, *Caryophyllaceæ* also would occur. Sufficient data relating to the affinities between *Aizoaceæ* and *Portulacaceæ* on the one hand, and *Caryophyllaceæ* on the other, not being available, we consider that inclusion of *Portulacaceæ* in the *Centrospermales* and the isolation of *Caryophyllaceæ* is not unjustified.

The classification modified on the above said lines would be as follows:

Centrospermeæ

Centrospermales	Chr. nos.	*Caryophyllales*	Chr. nos.
Chenopodiaceæ	3-series	*Caryophyllaceæ*	Aneuploid series from 8–13 and polyploid nos. in the 5-series
Amaranthaceæ	do.		
Nyctagineæ	29		
Phytolaccaceæ	3-series		
Aizoaceæ	3 & 4 series		
Portulacaceæ	do.		

5. *Summary*

The development of the microsporangium of *Portulaca tuberosa*, Roxb., is described. Multinucleate tapetal cells and tapetal nuclear fusion are met with.

The diploid number of chromosomes was found to be 18, which was confirmed by the meiotic counts. Diploid chromosome numbers of two other species of *Portulaca* are also recorded for the first time.

Meiosis is normal and the microspore is two-celled at the shedding time.

Development of the ovary and the ovules is described in some detail. Embryo-sac formation and embryo development are of the normal type.

The possible meaning of the tapetal nuclear fusion is discussed from the view-point of the nutritive function of the tapetal cells.

The importance of recounting morphological details in spite of their repetition, is discussed in the light of observations recorded in this paper. The position of *Portulacaceæ* is considered cyto-taxonomically.

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