Cytological Investigation of the Family Scrophulariaceae II. Coenocytism in *Digitalis purpurea* Linn.

R. Chandran and K. V. Bhavanandan

Department of Botany, University of Kerala,
Kariavattom, Trivandrum-695581, India

Accepted September 25, 1985

Occurrence of coenocytes or syncytes or multinucleate pollen mother cells (PMCs) is a rare phenomenon in angiosperms. However, their occurrence both spontaneous as well as induced has been reported in a few plant species, viz., *Aegilops squamosa* (Kihara and Lilienfeld, 1934), *Petunia* (Levan 1939, Nath and Nielsen 1961), *Phleum pratense* (Levan 1941), *Hordeum vulgare* (Smith 1942, Sethi *et al.* 1983), *Trillium erectum* (Stern 1946) *Phacellanthus* (Matsuura 1935), *Saccharum robustum* and *Erianthus arundinaceae* (Price 1956), *Digitaria eriantha* (Nath *et al.* 1970), *Triticale hexaploide* (Kempanna *et al.* 1971), *Saccharum bangalense* (Mehra and Kalia 1973) and *Zea mays* (Golubovskaya and Mashnenkov 1977). The present paper deals with the spontaneous occurrence of coenocytism in *Digitalis purpurea* from Ootacamund, South India.

Materials and methods

The materials of *D. purpurea*, existing in three forms (purple, white and yellow-flowered) were collected from two high altitude regions in the Nilgiris, namely Ootacamund and Kodaikanal. Young flower buds were fixed in 3:1 alcohol-acetic acid between 9.30 and 10.30 AM and squashed in 2% acetocarmine. A fixation period of three to five days and addition of few drops of ferric acetate solution to the fixative one day prior to squashing yielded better results. Pollen sterility was determined on the basis of stainability using acetocarmine. Photomicrographs were made from temporary slides.

Observation

All the forms of *D. purpurea* are morphologically identical except for the flower colour. The white and yellow-flowered forms from Ootacamund and purple-flowered forms from Kodaikanal showed 28 bivalents in the PMCs at diakinesis followed by regular course of meiosis resulting in appreciable pollen fertility of 94%, 93% and 97% respectively (Figs. 1, 2, 3), but the purple-flowered material from Ootacamund showed 38% of coenocytic PMCs and the rest normal PMCs. Two types of coenocytic PMCs were recognized in them such as 1) PMCs (78%) possessing 2-14 nuclei of similar size distributed in the common cytoplasm (Fig. 4) and 2) those (22%) having a single giant nucleus. Meiotic division in the multinucleate PMCs was either synchronous (53%) or asynchronous (47%). In the former case all the nuclei in the cytoplasm underwent meiotic division simultaneously so that identical stages were seen (Fig. 5). But in the latter the different nuclei in the coenocyte showed different meiotic stages (Figs. 6, 7). The coenocytes with single giant nucleus, however, showed multivalents, bivalents and univalents at the same metaphase I plate (Fig. 8). In spite of appreciable frequency of normal PMCs undergoing regular meiosis, the plant showed complete pollen sterility (Fig. 9).

Discussion

Cytological investigation in the purple-flowered materials of *D. purpurea* collected from Ootacamund was characterised by the occurrence of notable proportion of coenocytic PMCs. The purple-flowered material from Kodaikanal as well as the white and yellow-flowered forms collected from Ootacamund, however, showed normal PMCs with 28 bivalents. Comparable cases of coenocytism has been previously reported in a few other plants such as species of *Petunia* and *Phleum* (Levan 1939, 1941) in which such coenocytes were considered to have arisen by fusion of 2 to 30 PMCs, formed by the fusion of adjacent cells on the dissolution of intercellular walls. This was later substantiated by many other investigators (Matsuura 1935, Price 1956, Mehra and Kalia 1973, Sethi *et al.* 1983). But some others (Price 1956, Rees 1957, 1961, Kempanna *et al.* 1971) have considered the coenocytism having been arisen by the failure of cytokinesis in the mitotic division preceding meiosis. Based on the experimental evidence in *Trillium erectum*, Stern (1946) has pointed out surface tension and protoplasmic consistency

Figs. 1-9. *Digitalis purpurea*, purple-flowered strain from Kodaikanal. All figs. except Figs. 3 and 9. $\times 950$. 1, PMC showing 28 bivalents at metaphase I. 2, PMC at telophase II. 3, fertile pollen grains. $\times 150$. 4-9: *Digitalis purpurea*, purple-flowered strain from Ootacamund. 4, coenocyte showing five nuclei. 5, coenocyte with six nuclei showing synchronous division. 6, 7, coenocytes showing asynchronous division. 8, coenocyte showing multivalents, bivalents and univalents arranged in the same plate. 9, sterile pollen grains. $\times 150$.

as the physical properties governing the fusion of PMCs. Sethi *et al.* (1983) reported the occurrence of both spontaneous as well as induced coenocytism by colchicine and radiation and according to them coenocytes are formed by the fusion of PMCs at different stages of meiosis.

In the present material the coenocytic phenomenon has occurred spontaneously and were visible right from the early stages of meiosis. This appears to indicate that the coenocytes might have formed possibly by the fusion of adjacent cells on the dissolution of intercellular walls. Synchronous meiotic division was noticed in those coenocytes which formed out of like-aged PMCs. But in PMCs in which there was asynchronous meiotic division, fusion of differently aged PMCs might have taken place.

Various explanations have been proposed to account for the incidence of coenocytism in angiosperms. According to Smith (1942), Rees (1957) and Nielsen (1960) coenocytic formation is controlled by a recessive factor. More recently Kempanna *et al.* (1971) held that the arrest of cell wall formation after karyokinesis at premeiotic stages might be due to the functions of a gene or a gene system. A few others, however, reported coenocytism to be the result of mutation effects, such as pam-A 344 induced by N-nitroso-N-methylurea in maize by Golubovskaya and Mashnenkov (1977) and gamma radiations in barley by Sethi *et al.* (1983).

In the present material, the phenomenon of coenocytism has been noticed only in the purple-flowered strain occurring in Ootacamund in which this occurs consistently. However, purple-flowered strains growing in the climatically and altitudinally similar Kodaikanal hills of South India have not been found to exhibit the type of coenocytism. This appears to indicate that the phenomenon is not the result of any environmental effect. Possibly the coenocytic phenomenon occurring in the Ootacamund material could be due to natural mutations as has earlier been reported in a few other angiosperm taxa.

Summary

Spontaneous occurrence of coenocytism has been reported in the PMCs of purple-flowered strain of *Digitalis purpurea* from Ootacamund. Report of this kind is new not only for the genus but also for the family Scrophulariaceae. This phenomenon was visible right from the early stages of meiosis. Both synchronous as well as asynchronous nuclear division in coenocytes has been observed depending upon the stage of the PMCs taking part in fusion. The coenocytes are larger than the normal PMCs depending upon the number of PMCs involved in fusion. It is assumed that coenocytes were formed by the fusion of adjacent PMCs on dissolution of intercellular walls. It is also concluded that coenocytism in the present material could be due to natural mutations.

Acknowledgements

The authors wish to express their gratitude to Prof. (Dr.) C. A. Ninan, Head of the Department of Botany, University of Kerala for providing necessary facilities and encouragement. We are also grateful to Prof. P. M. Mathew for critical reading of the manuscript. Thanks are also due to Mr. A. Krishnankutty Nair for assistance in photographic work.

References

- Golubovskaya, I. N. and Mashnenkov, A. S. 1977. Multiple disturbances at meiosis in maize caused by a single recessive mutation pam A-A344. *Genetika, U.S.S.R.* **13**: 1910-1921 (Ru).
- Kempanna, C., Seetharam, A. and Shiva Shankar, G. 1971. Multinucleate pollen mother cells in Triticales. *Curr. Sci.* **49**: 301-302.

- Kihara, H. and Lilienfeld, F. 1934. Hereinwanderung und Bildung syndiploider Pollenmutterzellen bei dem F_1 -Bastard, *Triticum aegilopoides* und *Aegilops squarrosa*, Japan. J. Genet. **10**: 1-28.
- Levan, A. 1939. Tetraploidy and octoploidy induced by Colchicine in diploid *Petunia*. Hereditas **25**: 109-131.
- 1941. Syncyte formation in the pollen mother cells of haploid *Phleum pratense*. Hereditas **27**: 243-253.
- Matsuura, 1935.. c. f. Maheshwari, P. 1950. An Introduction to the Embryology of Angiosperms. p. 41, McGraw-Hill Book Company Inc.
- Mehra, P. N. and Kalia, V. 1973. Accessory chromosomes and multinucleate pollen mother cells in *Saccharum bengalense* Retz. Complex. Nucleus **26**: 75-88.
- Nath, J. and Nielsen, E. L. 1961. Cytology of plants from self and open-pollination of *Phleum pratense*. Amer. J. Bot. **48**: 772-777.
- , Swaminathan, M. S. and Mehra, K. L. 1970. Cytological studies in the tribe Paniceae, Gramineae. Cytologia **35**: 111-131.
- Nielsen, E. L. 1960. Cytology and fertility of 'small plants' in *Bromus enermis*. Bot. Gaz. **121**: 133-139.
- Price, S. 1956. Cytological studies in *Saccharum* and allied genera I. Syncytes in certain clones of *Saccharum* and *Erianthus*. Cytologia **21**: 21-37.
- Rees, H. 1957. The genotypic control of chromosome behaviour in rye IV. The origin of new variation. Heredity **11**: 185-193.
- 1961. Genotypic control of chromosome form and behaviour. Bot. Rev. **27**: 288-318.
- Sethi, G. S., Kalia, V. and Negi, M. 1983. Cytogenetics of coenocytism in barley (*Hordeum vulgare* L.) Cytologia **48**: 511-517.
- Smith, M. 1942. Cytogenetics of a factor for multiploid sporocytes in barley. Amer. J. Bot. **29**: 541-556.
- Stern, H. 1946. Formation of polynucleated pollen mother cells in *Trillium erectum*. J. Heredity **37**: 46-50.
-