ISSN 0376-5561
Geophytology 44(1): 87-90
May 2014

Abortive embryonic structures within megaspores of *Isoetes* L. (Lycopsida - Pteridophyta)

B. D. Sharma[1]*, D. R. Bohra[2], O. P. Suthar[3], R. Harsh[4] and S. N. Purohit[5]

[1]Kath Mandi, Narnaul-123001, India
[2]4B/7 Partap Nagar, Jodhpur-342001, India
[3]Botany Department, Government Girls College, Balotra-344022, India
[4]Botany Department, M.S. Government Girls College, Bikaner-334001, India
[5]E-110 1st Extension, Kamla Nehru Nagar, Jodhpur-342001, India
E-mail: bdsharma14@yahoo.com; drbohra14@gmail.com; opsuthar59@yahoo.co.in; rharshpalaeobotanist@yahoo.com; snpurohit983@gmail.com
*Corresponding author

Manuscript received: 03 January 2014
Accepted for publication: 10 March 2014

ABSTRACT

Sharma B. D., Bohra D. R., Suthar O. P., Harsh R. & Purohit S. N. 2014. Abortive embryonic structures within megaspores of *Isoetes* L. (Lycopsida–Pteridophyta). Geophytology 44(1): 87-90.

Isoetes L. is a herbaceous, aquatic to terrestrial lycopod in which mega and microsporangia are produced on separate sporophylls (leaves). Megaspores are large and trilete whereas microspores are small and monolete (in majority). Thin serial sections through the megasporangial portion show presence of abnormal embryonic structures in the spore contents of the functional megaspores. Sterile spores do not have spore contents and are empty. This type of abnormal embryos have been reported for the first time in the genus *Isoetes* L.

Key-words: Heterosporous quillwort, in situ abnormal embryogeny, Rajasthan, India.

INTRODUCTION

Isoetes L. (Isoetaceae) is a herbaceous, aquatic, amphibious or terrestrial green plant with a reduced lobed stem in the form of a corm or rhizomorph. Roots are forked and originate in regular rows. Leaves are ligulate and acicular and are produced in close spirals. Leaf size ranges from 4-5 cm (*I. reticulata*) to 60-70 cm (*I. coromandelina, I. tuberculata*) (Gena & Bhardwaja 1984, Sharma et al. 1985b). A sessile mega- or microsporangium is produced in the basal adaxial portion of every leaf. In some species, viz. *I. coromandelina* and *I. pantii*, mixed bisexual sporangia are also rarely reported (Goswami & Arya 1970, Singh et al. 1984, Goswami & Bhu 1998). A megasporangium has 200-600 or more trilete megaspores which are of two types, i.e. fertile (functional) and sterile (non-functional). The sterile spores are of various types, i.e. smaller spores, bigger spores, jointed spores, fused spores and enucleated spores (Verma 1960, 1961, Pant & Srivastava 1962, Bohra et al. 1980, Panigrahi 1981, Sharma et al. 1985b, 2008, Sharma & Purohit 2013). Ekambaram and Venkatanathan (1933) studied sporogenesis of *I. coromandelina* and reported 50% fertile and 50% sterile spores in a tetrad. Pant and Srivastava (1965) favoured this opinion. But Sharma et al. (1985a) reported variations in this ratio of fertile and sterile spores. A megaspore has three walls; the outer wall (exine) is thick and irregular due to the presence of ornamentations (tuberculate, reticulate, spinose, etc); the middle wall is thick,

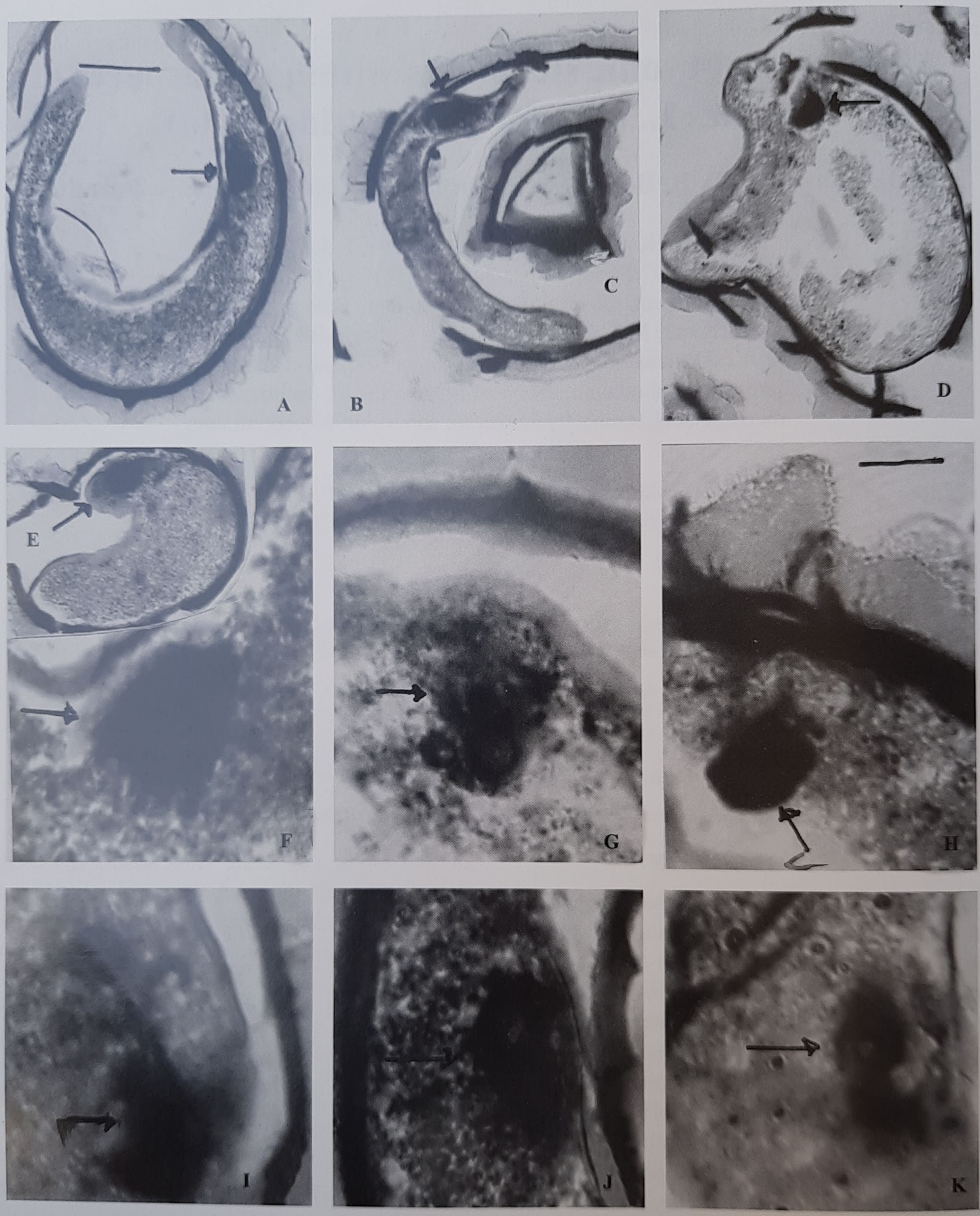

Plate 1

A–K. Sections through the megaspores of *Isoetes* sp. containing apogamous abortive embryos. A, B, D, E. Various shapes of spore contents which have isolated nucleated cells and large dark coloured (arrow) embryonic structures. C. Empty sterile spore with three wall layers. F. Embryonic structures enlarged, finger like outgrowth present. G. Cavities present in the embryo. H. Embryo with two small outgrowths. I. A small globular embryo and the larger curved one having a hook like outgrowth. J. Laterally elongated embryo with a cavity. K. Embryo with a cavity. (Bar: A–E = 75 μm, F–K = 30 μm).

hard and dark brown while the inner wall is thin and encloses the spore content.

Goebel (1897), for the first time, reported existence of apogamy in *Isoetes*. Ninan (1958) noticed apogamy in South Indian species of *Isoetes*. Pant and Srivastava (1965) observed apomictic germination in some Indian species of *Isoetes*. Srivastava et al. (1996) noticed in situ germination of megaspores in *I. panchganiensis* and *I. mahadevensis*. The present investigation is based on the study of thin serial sections through megasporangia of four species of *Isoetes*, viz. *I. coromandelina* L.F., *I. tuberculata* Gena & Bhardwaja 1984, *I. reticulata* Gena & Bhardwaja 1984 and *I. rajasthanensis* Gena & Bhardwaja 1984, found in Rajasthan, India. All these species have shown more or less identical results.

MATERIAL AND METHOD

The material of the four species (mentioned above) of *Isoetes* was collected from different places, viz. Mt. Abu, Daosa, Atru, Jhalawar, Chittorgarh, Menal, etc. Megasporangial portions of young and mature plants were separated and fixed in F.A.A. (formalin, acetic acid and alcohol) and then preserved in 70% alcohol. The material was then processed for microtomy as suggested by Johansen (1940). Sections were stained by the combination of safranin and haemotoxylin and mounted in dilute canada balsam.

OBSERVATION AND DISCUSSION

In some of the megaspores, the entire circular space is occupied by the spore content while in others, it reduces to U-shaped (Plate 1, figure A), C-shaped (Plate 1, figures B, E) or a triarch structure. In the circular spore content, a small cavity appears which increases in size and changes the shape of the spore content to U, C or triarch structure. This change in shape and size also depends on the plane and position of sectioning of the megaspore. In the spore content develops free floating enucleated and nucleated cells (Plate 1, figure K). The latter are circular with distinct dark coloured nuclei. These are isolated as well as in group of 4-5 or more cells. In the spore content of some of the cells, large sized dark coloured bodies are also seen (Plate 1, figures A, B, D, E). These are globular, laterally elongated or irregular in shape (Plate 1, figures F-K). Their position in the spore content is also variable, i.e. deep in the spore content, along the outer periphery or inner periphery or terminal. These embryonic bodies originated from the nucleated cells of the megagametophytes and are of various shapes and sizes (Plate 1, figures F-K). In many of these bodies, circular or elongated cavity or cavities are visible (Plate 1, figures G, J, K) identical to those drawn by Hofmeister (1862, plate 46) in his description of development of embryo in *Isoetes*. We call the bodies present in our material as embryos or embryonic structures. Archegonia are not seen in any megaspore. The embryonic structures are apogamous for want of microspores (or spermatozoids) and fertilization. These are abortive because of their origin directly from the free floating nucleated cells of the megagametophyte present in the spore contents that is, not protected by archegonia. Differentiation of organs, like foot, root, leaves, etc., is not visible in the embryonic structures. In one or two, a long root like structure has been seen. Similarly, a curved hook like structure (Plate 1, figure I), probably a leaf primordium, is visible in few embryos. In some ferns, like *Adiantum lunulatum*, archegonia are absent and all cells of the prothallus are capable to form an apogamous sporophyte (Mehra & Gupta 1986, Sharma & Sharma 1992). A similar condition has been observed in *Isoetes*. The free floating nucleated megagametophytic cells form apogamic embryonic structures. Further investigations are required on these embryonic bodies on their mode of origin and behaviour.

ACKNOWLEDGEMENT

The authors express their sincere thanks to Dr. Ravinder Singh for preparation of slides and MS Surya Shekhawat for assistance in photography.

REFERENCES

Bohra D. R., Sharma B. D. & Singh R. 1980. *Isoetes* in Rajasthan - A study of spores. Adv. Pollen Res. 5: 1-8.

Ekambaram T. & Venkatanathan T. N. 1933. Studies on *Isoetes coromandelina* L. I. Sporogenesis. J. Indian Bot. Soc. 12: 191-225.

Gena C. B. & Bhardwaja T. N. 1984. Three new species of *Isoetes* L. from Rajasthan. J. Bombay Nat. Hist. Soc. 81: 165-168.

Goebel K. Von 1897. Ueber sprossbildung auf *Isoetes* blatten. Bot. Zeit. 37: 1-6.

Goswami H. K. & Arya B. S. 1970. A new species of *Isoetes* from Narsinghgarh, Madhya Pradesh. J. Indian Bot. Soc. 49: 30-37.

Goswami H. K. & Bhu I. 1998. Bisexual sporangia in *Isoetes pantii*. Bionature 18: 31-36.

Hofmeister W. 1862. On the germination, development and fructification of higher cryptogamia and fructification of the Coniferae. Roy. Soc. London.

Johansen D. A. 1940. Plant Microtechnique. McGraw Hill, New York.

Mehra P. N. & Gupta A. 1986. Gametophytes of Himalayan ferns. Botany Department, Panjab Univ., Chandigarh, India: 1-204.

Ninan C. A. 1958. Studies on the cytology and phylogeny of pteridophytes. V. Observations on Isoetaceae. J. Indian Bot. Soc. 37: 93-103.

Panigrahi G. 1981. Systematics of the genus *Isoetes* L. (Isoetaceae) in India. Biol. Mem. 6(2): 129-138.

Pant D. D. & Srivastava G. K. 1962. The genus *Isoetes* in India. Proc. Natl. Inst. Sci. India 28B: 242-280.

Pant D. D. & Srivastava G. K. 1965. Cytology and reproduction of some Indian species of *Isoetes*. Cryologia 30: 239-251.

Sharma A. Sharma B. D. 1992. Gametophytes of five species of Rajasthan ferns under cultural conditions. Bull. Bot. Surv. India 32: 151-161.

Sharma B. D., Bohra D. R., Suthar O. P. & Harsh R. 2008. A brief account of history and various aspects of researchers on *Isoetes* Linn. Indian Fern J. 25: 167-201.

Sharma B. D., Bohra D. R. & Vyas M. S. 1985a. Sporogenesis in *Isoetes*. Bionature 5: 9-13.

Sharma B. D. & Purohit S. N. 2013. Spores, germination and apogamy in *Isoetes* of Rajasthan, India. Geophytology 43(2): 177-182.

Sharma B. D., Singh R. & Vyas M. S. 1985b. *Isoetes* in Rajasthan – A systematic study. Bull. Bot. Surv. India 27: 58-62.

Singh R., Bohra D. R. & Sharma B. D. 1984. Mixed sporangia in *Isoetes coromandelina* L. Pollen Spores 25: 41-47.

Srivastava G. K., Shukla P. K., Wagai S. O. & Srivastava M. 1996. In situ germination of megaspores of some Indian species of *Isoetes* L. Natl. Acad. Sci. Letters 19: 191-194.

Verma S. C. 1960. Enucleate spores in *Isoetes coromandelina* L. Caryologia 13: 274-284.

Verma S. C. 1961. Cytology of *Isoetes coromandelina* L. Amer. Fern J. 51: 99-104.