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Phyton (Austria)	Vol. 14	Fasc. 1–2	165–173	16. XII. 1970

Contribution from the Department of Botany, University of Lucknow, Lucknow (India), New Series (Bryophyta) No. 65

Sporeling Development in *Preissia quadrata*

By

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With 40 Figures

Received August 31, 1970

Introduction

INOUE 1960 made exhaustive study of spore germination in various members of the *Marchantiales*. His studies include the process of spore germination and development of young gametophyte under different environmental conditions. He observed that environmental factors do not influence the pattern of spore coat dehiscence which is more or less stabilized and characteristic for various taxa. However, the germ rhizoid formation which is also sufficiently distinctive may often get modified under different conditions.

Inspite of several contributions on the spore germination in *Marchantiales* by the earlier workers viz. HEDWIG 1784, MIRBEL 1836, BISCHOFF 1853, GROENLAND 1845, GOEBEL 1930, O'HANLON 1926, 1927, 1930, DUTHIE & GARSIDE 1937, 1940, MEHRA & KACHROO 1951, 1952, UDAR 1958a, 1958b, UDAR & SRIVASTAVA 1968, JOVET-AST 1963 and various others, there still remain some taxa which have not been worked out in sufficient details. Even some of the species already worked out elicit desirability for reinvestigation for greater information. Studies on the sporeling development in hepatics have acquired a great significance in view of the fact that the observations have been of great help in understanding the evolutionary sequence and interrelationships of the various groups (INOUE 1960; FULFORD 1956; NEHIRA 1966). Consequently detailed study of such taxa which have

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been inadequately worked out or not investigated so far assumes considerable significance.

The present communication attempts to give a detailed account of sporeling development in *Preissia quadrata*, which needed reinvestigation due to inadequate details available from contributions by earlier workers (GROENLAND 1845; SCHOSTAKOWITSCH 1894; O'HANLON 1927 and INOUE 1960). Our study of sporeling stages in this genus provided some interesting results not encountered earlier.

Spore morphology of *Preissia quadrata*

The mature spores of *P. quadrata* are more or less rounded or triangular, brown, 43.2—55.2 (—52.6) μ in diameter, unicellular, cryptopolar and tetrahedral with inconspicuous tri-radiate mark on the proximal face (Fig. 2). The perinium is conspicuous and presents several folds towards the periphery. The distal face of the spore (Fig. 1) shows irregularly distributed fine lamellae.

Materials and methods

The fertile plants of *Preissia quadrata* were collected by one of us (UDAR) from Kashmir (On way to Sonamarg Glacier, ca. 11,000 ft.) during the month of October, 1968. The thalli had well developed stalked sexual receptacles borne terminally. The mature sporophytes were dissected out and subjected to repeated washing by distilled water. The spores were cultured twice: On 29th October, 1968 and on February 3, 1969: in sterilized covered pyrex glass petridishes containing:

a) Half strength Knop's Agar medium.

b) Full strength Knop's solution.

c) Half strength Knop's solution.

The Knop's solution as well as the Knop's Agar were prepared by the usual method. Difco Agar was used for preparing the Agar medium. The cultures received sunlight through North glass window panes of the laboratory at room temperature.

Observations

The spores cultured on October 29, 1968 germinated within four or five days (i. e. on November, 3, 1968) and those cultured on February 3, showed initial stages of germination on the seventh day (i. e. on February 9, 1969). This suggests that there is no rest period. About 90% of the spores had the persistent viability for at least about five months as evident by the present investigation. However, O'HANLON 1927 showed that about 10% of the spores had the persistent viability between 4 and 5 months. In the initial stages of spore germination there is the appearance of a germ

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rhizoid (Figs. 3—5) as in *Targionia hypophylla*, *Sauteria yatsuensis* (INOUE 1960) and *Athalamia pinguis* (UDAR 1958b). However, in some of the sporelings the germ rhizoid formation was slightly delayed (Figs. 29, 33), while few could not develop the germ rhizoid even at a late stage (Fig. 32). Subsequently a germ tube emerges through the proximal face of the spore (Fig. 7).

In most of the spores the germ cell, after getting enlarged by the absorption of water, divides into two unequal cells at a very early stage, by a transverse or more or less oblique wall (Fig. 6), resulting in the formation of a smaller cell which grows out to form a germ rhizoid and a larger cell which may develop into a germ tube or undergo further cell divisions forming a multicellular parenchymatous mass (Figs. 17—21) — a feature common in *Targionia hypophylla* (INOUE 1960) and *Athalamia pinguis* (UDAR 1958b).

However, in few cases the germ cell does not undergo any division and the germ rhizoid is formed as a protrusion of the germ cell itself (Figs. 3—5). In such sporelings where the germ rhizoid has an internal continuity with the germ cell, usually germ tube formation takes place (Figs. 7—16) while in those sporelings, where the germ rhizoid is separated by means of a wall, cell mass formation was commonly prevalent (Figs. 6, 18—22). Abnormally, however, a sporeling may show the formation of a germ tube as well as separation of germ rhizoid by a wall (Fig. 36).

The germ tube divides in two cells by a transverse septum (Fig. 8). The upper cell of the filament next divides in two cells by a vertical wall (Fig. 9) more or less at right angle to the first transverse wall. Subsequently each of the resulting cells divide transversely (Fig. 10) forming a four celled germ plate or germ disc (Fig. 10) as in most of the *Marchantiales*. An apical cell is subsequently organised by the activity of which a multicellular germ disc is formed (Figs. 11—16). In certain cases a filament of cells is formed by repeated transverse divisions (Fig. 36).

Besides the formation of germ disc from a germ tube, there were a number of sporelings in the same culture which showed the formation of a cell mass or occasionally stages with highly reduced germ tube. These sporelings were later observed to possess regeneration capacity (Figs. 21, 30—35, 38, 39). One of the cells of the germ disc in these sporelings protrudes out into a tube like structure (Figs. 30—31) which gets divided transversely into two (Figs. 32, 33) or sometimes into three cells (Fig. 35). The terminal cell of the above, divides again by a vertical wall to form two cells (Figs. 34, 35). Here the divisions are more or less in the same way as in the terminal cell of the filament (Fig. 9). Subsequent laying down of the transverse and longitudinal walls result in the formation of another germ disc (secondary germ disc) which remains attached to the previous one (Figs. 28, 35, 38, 39). Regeneration capability has also been observed in some of the germ discs which developed after the germ tube stage (Fig. 16).

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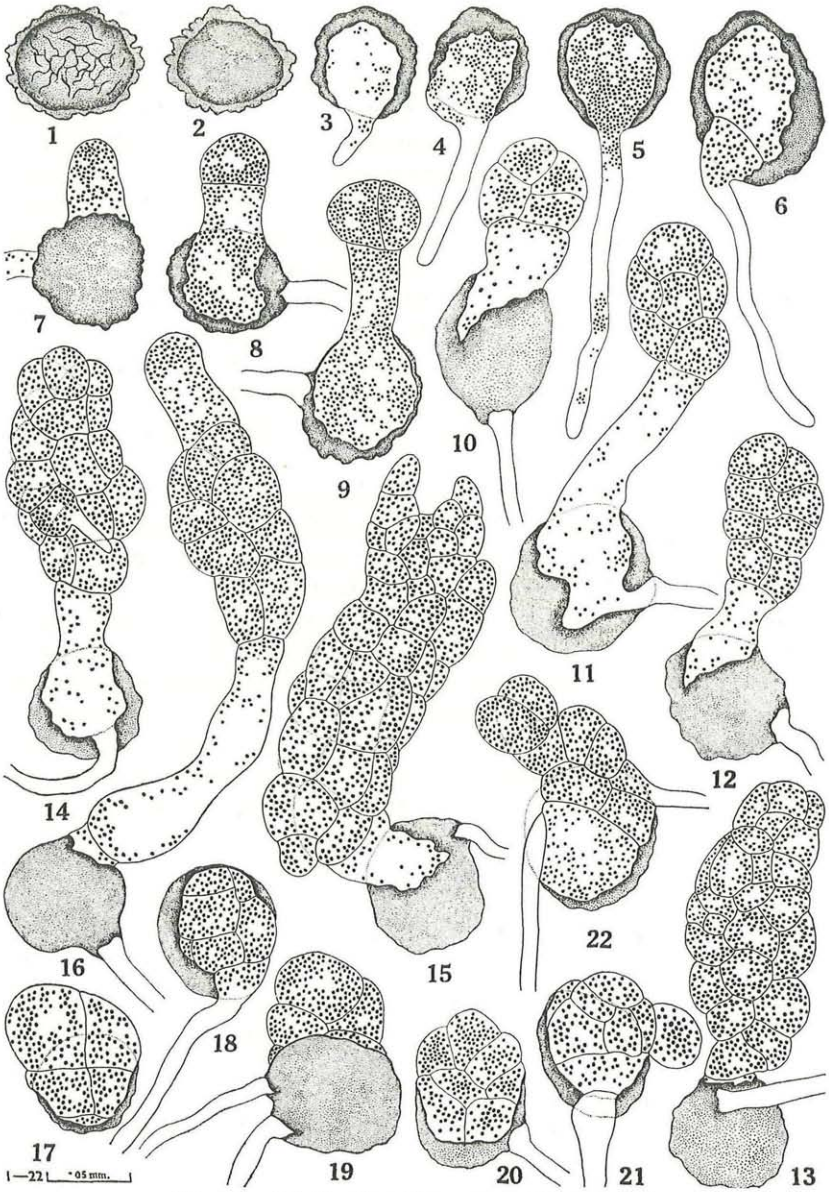

Figs. 1—22. Explanation see p. 168 and 169

Figs. 1, 2. Distal and proximal views of the spore respectively. — Figs. 3—5. Emergence of germ rhizoid. Note the continuity of the germ rhizoid with that of the germ cell. — Fig. 6. Emerging germ rhizoid being separated by means of an oblique septum. — Fig. 7. Formation of the germ tube and the germ rhizoid. — Fig. 8. Showing first transverse division of the germ tube. — Fig. 9. Appearance of a vertical wall at right angle to the first transverse wall. —

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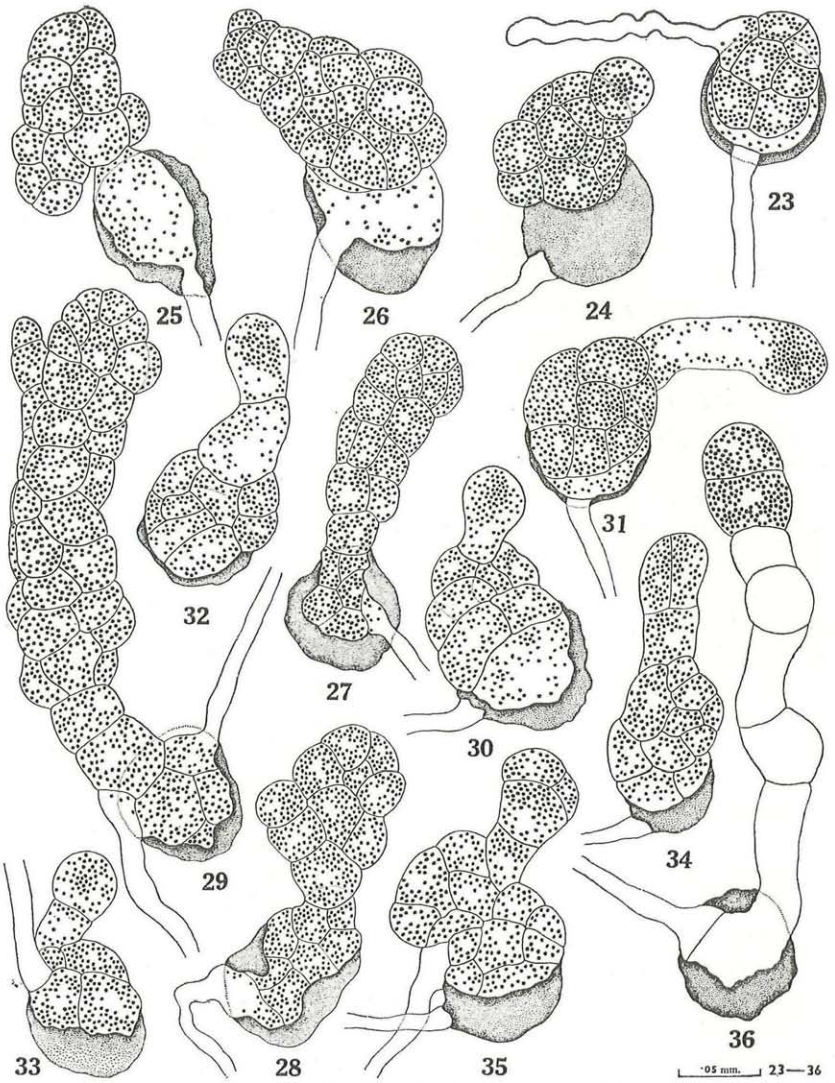

Figs. 23—36. Explanation see p. 169

Fig. 10. Four-celled germ disc. — Figs. 11—15. Subsequent stages in the development of the germ disc. Note the emergence of secondary rhizoid in Fig. 14. — Fig. 16. Formation of regeneration tube from a multicellular germ disc. — Figs. 17—24. Stages in the development of a cell-mass. — Figs. 25, 26. Sporelings with germ disc developing from highly reduced germ tube. — Figs. 27—29. Older stages of cell mass development. — Figs. 30—35. Stages of regeneration from cell mass. Note the appearance transverse wall (Figs. 32, 33) and subsequent vertical and transverse walls (Figs. 34, 35) in the regenerant arising from the cell mass. — Fig. 36. An abnormal filamentous stage showing the separation of germ rhizoid by means of a vertical wall.

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Conclusions

Like several other members of the *Marchantiales* (e. g. *Targionia hypophylla*, *Wiesnerella denudata*, *Marchantia tosana*, *M. cuneiloba*, *Monoselenium tenerum*, *Neohodgsonia mirabilis*), the present species (*Preissia quadrata*) has also cryptopolar spores. The spore coat dehiscence in all such species is through the proximal face except in *Targionia hypophylla* which shows irregular spore coat dehiscence (INOUE 1960). Besides this, the members of the family *Rebouliaceae* with polar spores (*Cryptomitrium himalayense*: KACHROO 1955; *Plagiochasma intermedium*: UDAR & CHANDRA 1965; *P. appendiculatum*, *P. articulatum*, *Mannia indica*, *Asterella blumeana*, *A. reticulata*, *A. mussooriensis* and *A. angusta*: MEHRA & KACHROO 1952) are also grouped under the same category with proximal spore coat dehiscence.

In majority of *Marchantiales* after the spore coat ruptures, the first stage of germination is the appearance of germ papilla (as in *Riccia* spp., members of the families *Rebouliaceae*, *Exormothecaceae* and *Sauteriaceae* except *Athalamia pinguis*). In these, the germ rhizoid formation is usually delayed. However, in *Preissia* quadrata, as also in *Marchantia polymorpha* (*Marchantiaceae*) and *Athalamia pinguis* (*Sauteriaceae*), the germ rhizoid formation precedes germ disc or germ tube formation. Under similar conditions of growth two types of germ rhizoid formation in *Preissia quadrata* is interesting: (1) the germ cell itself grows directly to form a germ rhizoid without any septum, sometimes containing chloroplasts (Figs. 3—5) which disintegrate afterwards and (2) the germ cell divides in two unequal cells by an oblique or transverse wall with the lower smaller cell protruding out to form the germ rhizoid (Fig. 6). INOUE 1960 treats germ rhizoid formation in *Preissia quadrata* under '*Reboulia*-type' in which the germ cell divides by a vertical wall forming two unequal cells, the smaller cell of which protrudes out to form a germ rhizoid parallel to that of the germ tube. The present study has however, shown that the germ rhizoid grows either opposite (Figs. 3—6, 10, 14, 18, 21, 23, 25, 26) or at right angle (Figs. 7—9, 11, 13, 19) or even very rarely more or less parallel (Fig. 40) to the germ tube or the germ disc. In some cases the germ cell divides by a transverse wall (Fig. 21) and the germ rhizoid grows opposite to germ disc corresponding to '*Marchantia*-type'.

The different types of germ rhizoid formation in *P. quadrata* show relationship to the germ disc formation at a later stage. It has been observed that the sporelings having germ rhizoid not separated by any septum (Figs. 3—5), show conspicuously developed germ tube (Fig. 7) which later organises the germ disc (Figs. 8—16), whereas the sporelings having germ rhizoid separated by a wall, more commonly develop directly into a cell mass (germ disc) and the germ tube formation in such cases is greatly reduced or absent (Figs. 6, 18—22, 24, 27, 31—35). However, Fig. 36,

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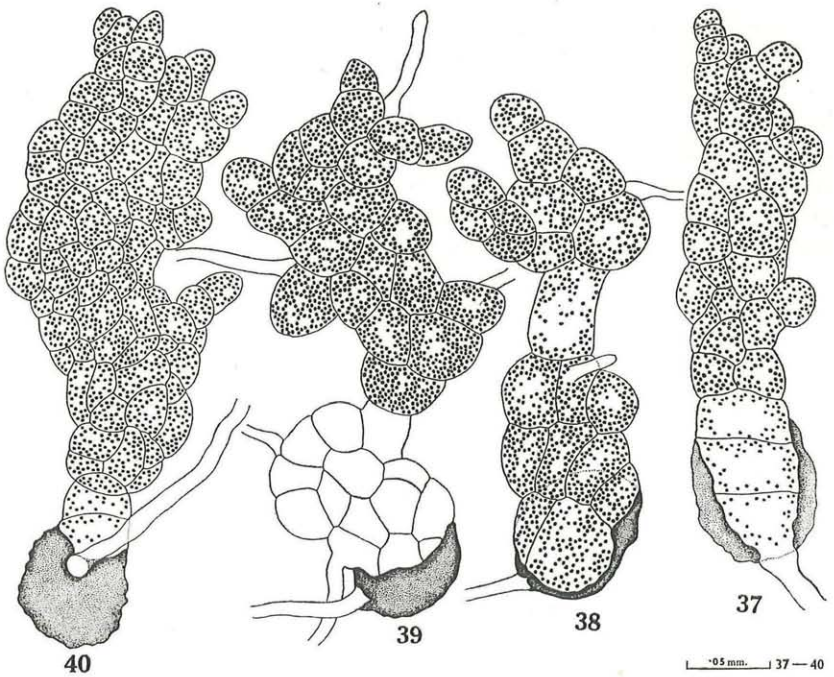
shows an abnormal filamentous stage where the germ rhizoid is already separated by an oblique vertical wall.

In sporelings developing through the formation of a germ tube different types of germ plate or germ disc formation take place. It may be eccentric (Figs. 15, 26) corresponding to '*Asterella*-type', or it may be in line with the germ tube (Fig. 12—14) corresponding to '*Stephensoniella*-type'. However, the formation of a cell mass occurs more frequently and is comparable to that in *Fossombronia cristula* (UDAR & SRIVASTAVA 1970),



Figs. 37—40. *Preissia quadrata*. Mature sporelings showing regenerants in Figs. 38 and 39.

Athalamia pinguis (UDAR 1958b) and *Targionia hypophylla*. But in species of *Fossombronia* the germ rhizoid formation is absent. Thus *P. quadrata* belonging to *Marchantiaceae* shows affinity with *Metzgeriales* on the one hand and with families *Rebouliaceae*, *Exormothecaceae*, *Sauteriaceae* and *Targioniaceae* on the other. The affinity of *Fossombronia* and *Preissia* in the formation of cell mass support MEHRA's 1957a, 1957b theory of origin of thallus structure in *Marchantiales*.

The stages of sporeling development in *P. quadrata* as outlined above reflect the flexibility in the germ rhizoid and germ disc formation. INOUE

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1960 remarked that the density of spores in culture medium in general, effects the growth of the sporeling. According to him when the spores are densely cultured the germ tube usually elongates and becomes several celled by the appearance of many transverse septa, whereas if the culture is monosporic the filamentous stage has been stated to be almost absent. The present investigation has clearly shown that even in the same culture the sporelings of various types i. e., with or without filamentous stage may develop.

Summary

The spore morphology and sporeling development in *Preissia quadrata* have been described. The germ tube emerges through the proximal face. The germ rhizoid formation is of two types which shows relationship with the germ disc formation at a later stage. The germ disc formation may be eccentric or in a straight line with the germ tube. The sporeling developmental stages in *Preissia quadrata* show affinity with *Metzgeriales* on the one hand and with the other members of the families *Marchantiaceae*, *Exormothecaceae*, *Rebouliaceae*, *Sauteriaceae* and *Targioniaceae* on the other.

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Zeitschrift/Journal: Phyton, Annales Rei Botanicae, Horn

Jahr/Year: 1970

Band/Volume: 14_1_2

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Artikel/Article: Sporeling Development in Preissia quadrata. 165-173