Timo KOPONEN* & Kunito NEHIRA**: The germination of asexual organs in the Mniaceae

知藻古保年（ティモ・コポネン)*・根平邦人**: チョウチンゴケ科における無性生殖器官の発芽

Although the specialized asexual organs, gemmae or propagula are rather common among the bryophytes (cf. Correns 1899), *Trachycystis flagellaris* was the only species within the Mniaceae known to have them, until the recent discovery of propagula in *Rhizomnium tuomikoskii* and *R. horikawae* (Koponen 1971 a; 1971 b). However, as stated by Correns (1899; cf. also Watson 1967) the separation of whole organs can be an effective means of spreading a plant. In the Mniaceae the good regenerating ability of detached stem parts and even detached leaves has been evidenced experimentally (Lersten 1961; Misiura 1964; Giles & von Maltzahn 1967) and observed in the field. The herbarium research has revealed that within the genus *Rhizomnium* many of the species have the ability to develop protonematous rhizoids which originate at the leaf axils. These have been checked for *R. punctatum*, *R. glabrescens* and *R. striatulum*. Moreover, most of the species of the *Rhizomnium* begin their growth by forming a dense protonematous rhizoid mat on moist rock surface or on rotten wood (Koponen 1971 a). The absence of the information on the developmental data of the asexual organs and the protonematous rhizoids seemed to call for a further research. This paper deals mainly with the germination of flagella, propagula and rhizoids and the development of the protonema. An attempt is made to discuss on the taxonomic significance of these organs.

Materials and methods

The materials used in this study are listed below. The voucher specimens are preserved in the Botanical Museum, University of Helsinki (H) and Herbarium of Hiroshima University (HIRO), or in the Hattori Botanical

* Department of Botany, University of Helsinki, Unioninк, 44, Helsinki 17, Finland.
** Biological Institute, College of General Education, Hiroshima University. 広島大学教養部生物学教室.

Laboratory (NICH).

Trachycystis flagellaris (Sull. et Lesq.) Lindle. (Koponen 19700, H, HIRO): The flagella were collected from Jumonji Pass, alt. 1770 m, Chichibu Mts., of Saitama Pref. on May 25, 1971 and sown on June 1, 1971.

Rhizomnium minutulum (Besch.) Kop. (Koponen 19623, H, HIRO): The protonematous rhizoids were collected from Jumonji Pass, alt. 1650 m, Chichibu Mts., of Saitama Pref. on May 25, 1971 and sown on June 1, 1971.

Rhizomnium striatulum (Mitt.) Kop. (Koponen 19969, H, HIRO): The protonematous rhizoids were collected from Ochigawa, alt. 650 m, Chichibu Mts., of Saitama Pref. on May 27, 1971 and sown on June, 1, 1971.

Rhizomnium tuomikoskii Kop. (Koponen 18864, H, HIRO): The propagula and rhizoids were collected from Miyajima, alt. 300 m, of Hiroshima Pref. on Jan. 15, 1971 and sown on Jan. 18, 1971.

The materials were sown on the liquid medium of one-half-strength

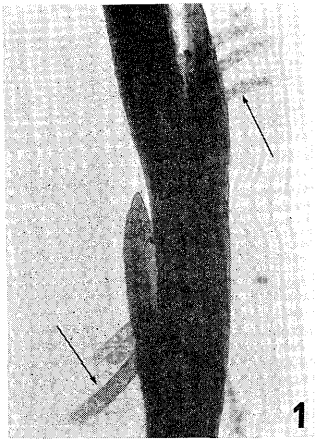


Fig. 1. Initial cells (arrows) originating from leaf axils of a flagellum of *Trachycystis flagellaris* ×100.

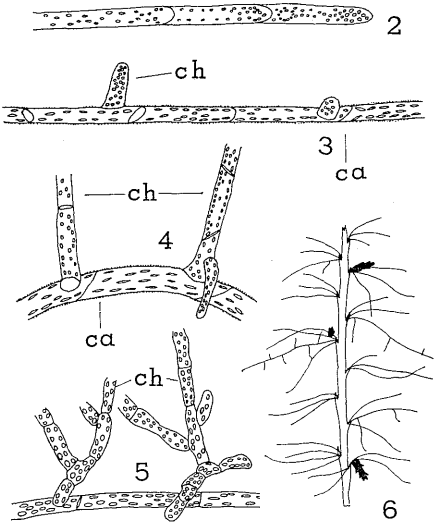


Fig. 2-6. *Trachycystis flagellaris*. 2. Apical part of the protonema developed from a flagellum ×130. 3, 4. Middle parts of protonema ×130. 5. Branched chloronema ×130. 6. Germinated flagellum. Leafy shoots seen on the basal part of caulonemata ×14. ca: caulonema, ch: chloronema.

Knop's solution in closed Petri dishes (6 cm in diameter) with about 10 ml of medium which was autoclaved before the experiment and kept in an incubator at 22°C–24°C under about 1000 luxes of continuous white light from fluorescent tubes. The medium was formulated as follows: $Ca(NO_3)_2 \cdot H_2O$(0.5 g), $MgSO_4 \cdot 7H_2O$(0.175 g), KH_2PO_4(0.175 g), KCl(0.06 g), $FeCl_3$ (trace) and distilled water (1000 ml). The pH of the medium was adjusted to 5.8.

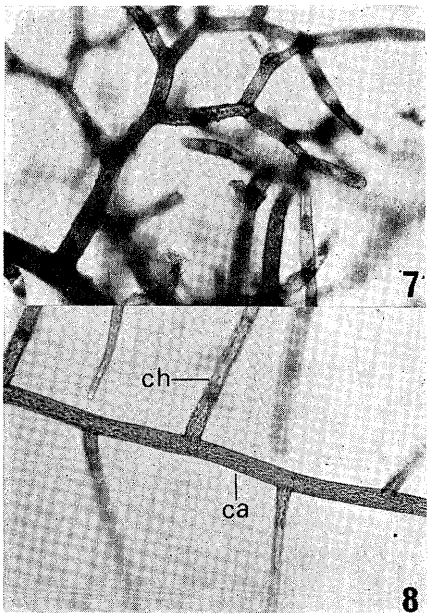
Rhizomnium horikawae (Nog.) Kop.*: The specimen used for photographing (Fig. 21): East Nepal. Baroya Khimty—Thakma Khola, alt. 2500–3000 m, Nov. 16, 1963, H. Kanai, G. Murata & M. Togashi (NICH).

Results and discussion

Trachycystis flagellaris: Correns (1899) described the structure and the burst of the flagella in *T. flagellaris*. He also observed the rhizoid-like structure at leaf axil of detached flagella in the herbarium specimens, but he did not describe it in detail. After one day's treatment, initial cells of the protonema are produced at leaf axils of the flagella (Fig. 1). The first septum is transversely formed at the elongated cell. By further transversal segmentations, a long filamentous protonema is developed. Later the septa seem to become oblique in every portion of the protonema. At about 10-celled stage of the filamentous protonema, chlorophyllous cells are branched from it. The protonema shows heterotrichous habit in the structure of strong dark-brown filaments with papillae of prostrate system (caulonema) and richly chlorophyllous aerial filaments (chloronema). The chloroplasts differentiate into three forms: granular in the apical part of the protonema (Fig. 2), spindle-like in the caulonema (Figs. 3, 4) and globular in the chloronema (Fig. 5). On the basal part of the caulonema, a pro-bud cell is formed and gives rise to the leafy shoot with many dark-brown rhizoids (Fig. 6).

Rhizomnium minutulum: The protonematous rhizoid of *R. minutulum* is composed of two parts; the dark-brown main filament bearing oblique septa and the greenish brown branches (Fig. 7). The initial cells are formed both on the main filament and on the branches. The initial cell immediately elongates and gives rise to the caulonema by further segmentations

* Thanks are due to Dr. S. Hattori of the Hattori Botanical Laboratory for loan the specimen of *R. horikawae*.



Figs. 7, 8. *Rhizomnium minutulum* 7. Protonematous rhizoid ×100. 8. Heterotrichous protonema ×100. ca: caulonema, ch: chloronema.

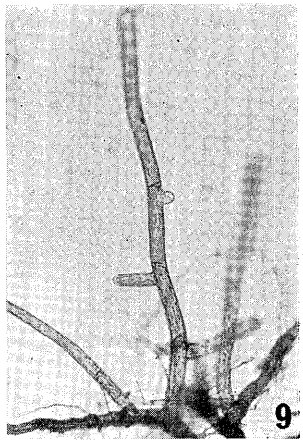


Fig. 9. Protonema developed from protonematous rhizoid of *Rhizomnium striatulum* ×100.

with oblique septa. The chlorophyllous cells are usually branched from the caulonema (Fig. 8). The chloronema is rarely formed directly at the middle part of the main filament (Fig. 10). Sometimes the apical cell of the rhizoid directly develops into caulonema (Fig. 11). As shown in Fig. 12, the protonema is of a typical heterotrichous habit consisting of prostrate caulonema and aerial chloronema. The caulonema has usually brown cell-wall, oblique septa and relatively deficient in chloroplasts. The chloronema of *R. minutulum* is richly chlorophyllous and more branched (Fig. 13) than that of the other *Rhizomnium* species studied by us.

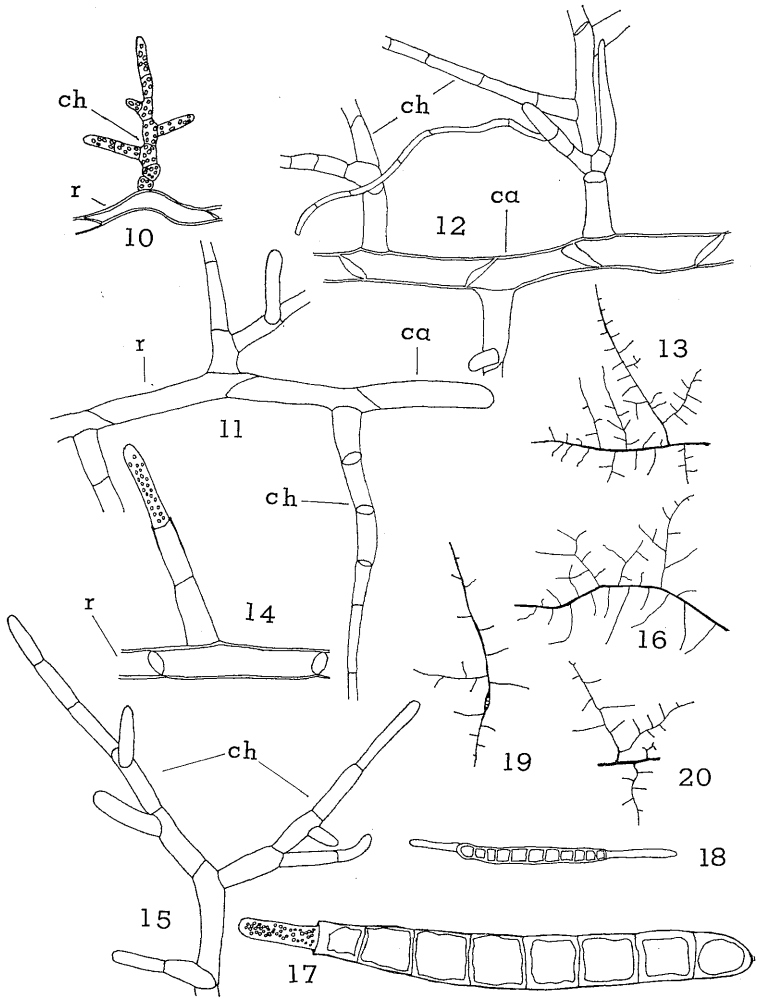
Rhizomnium striatulum: *R. striatulum* is rather similar to *R. minutulum* in the protonematous rhizoid structure and in the formation pattern of the protonema. Fig. 14 shows an initial cell originating from a cell of branched rhizoid. Later the filamentous caulonema branches frequently (Fig. 9). Sometimes the apical cell of the rhizoid directly develops into a chloronema

(Fig. 15). Fig. 16 shows a protonema grown up in cultivation.

Rhizomnium tuomikoskii: On the germination of the propagulum of *R. tuomikoskii*, an initial cell is produced both at the portion attached to rhizoids (Fig. 17) and at the opposite side (Fig. 18). The cell elongates and the first septum is formed at the elongated cell. By further transversal segmentations, the filamentous protonema is developed. All the septa of the protonema are rather oblique. Rhizoid is able to produce initial cells after the propagula have been detached. Then the initial cells of the protonema are produced at the apical cells of the stalk attached to propagula (Fig. 23). By further segmentations, the caulonema with oblique septa is formed as described for propagula. The chlorophyllous cells are developed on the caulonema originating from both propagula and rhizoids (Fig. 24). Figs. 19 and 20 show the cultivated protonemata originating from a propagulum and from a rhizoid respectively.

The propagula of *R. tuomikoskii* and *R. horikawae* are a valuable specific character which separetes the other *Rhizomnium* species. Moreover, as shown in Figs. 21 and 22, the propagula of *R. horikawae* are longer and more slender than those of *R. tuomikoskii*. This difference serves a character for separating these species from each other. The gemmae produced on the rhizoid system have previously been known from *Bryum erythrocarpum* and *Leptobryum pyriforme* (e.g. Watson 1967), in which the shape of the gemmae is nearly spherical and ovoid, respectively. The differences in the shape of the propagula has been used as a specific character in the propaguliferous species of the genus *Pohlia*.

In addition to that the propagula of *Rhizomnium tuomikoskii* and *R. horikawae* and the flagella of *Trachycystis flagellaris* are useful specific characters, these organs and the protonematous rhizoids described above may have a wider taxonomic significance. As mentioned above, the initial stage of most of the *Rhizomnium* species is a protonematous rhizoid mat. These organs have been observed in the nature for *R. glabrescens*, *R. punctatum*, *R. striatulum* and *R. minutulum* (Koponen 1971a) and checked also for *R. hattorii* and *R. perssonii*. A similar growth of rhizoids has not been seen in the other groups of the Mniaceae. Protonematous rhizoids originating from leaf axils on the stems of other genera than *Rhizomnium* has not been seen either. Thus, the macronematous rhizoids with a good regenerating



Figs. 10-13. *Rhizomnium minutulum*. 10. Chloronema originating from protonematous rhizoid ×130. 11. Apical part of rhizoid ×130. 12. Middle part of protonema ×130. 13. Protonema ×14.

Figs. 14-16. *Rhizomnium striatulum*. 14. An initial cell originating from a rhizoid ×130. 15. Apical part of branching chloronema ×130. 16. Protonema ×14.

Figs. 17-20. *Rhizomnium tuomikoskii*. 17. An initial cell originating from a propagulum ×130. 18. Germinated propagulum ×33. 19, 20. Protonemata developed from a propagulum and from a rhizoid respectively ×14. ca: caulonema, ch: chloronema, r: protonematous rhizoid.

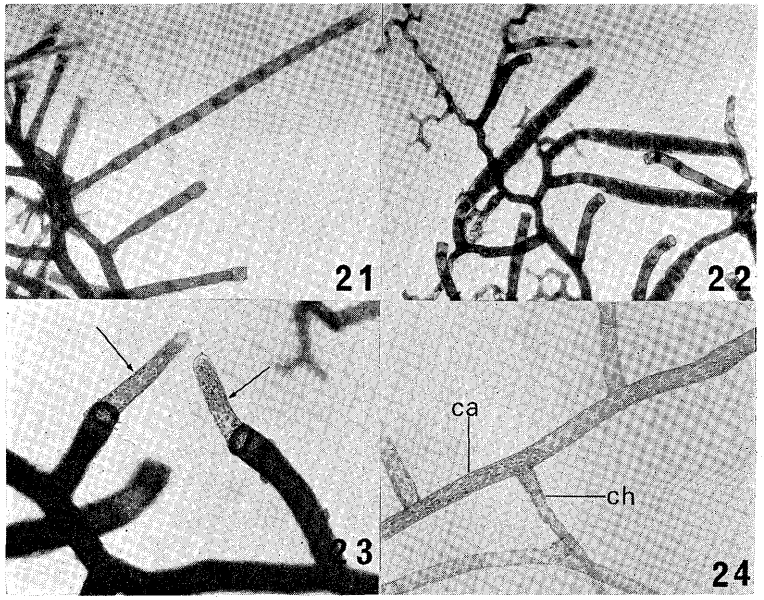


Fig. 21. Rhizoid of *Rhizomnium horikawae* ×40. Figs. 22–24. *Rhizomnium tuomikoskii*. 22. Rhizoid ×40. 23. Initial cells (arrows) originating from a rhizoid ×100. 24. Heterotrichous protonema developed from a rhizoid ×100. ca: caulonema, ch: chloronema.

ability and the presence of protonematous rhizoid mat in *Rhizomnium* possibly can be used as a generic character. This view is emphasized by the presence of propagula born on rhizoids in two of the species, *R. tuomikoskii* and *R. horikawae*.

The regenerating pattern of the flagella in *Trachycystis flagellaris* bears a special significance. According to Koponen (1968), the species of the tribes Mniae and Cinclidiae of the Mniaceae have only macronemata (except *Rhizomnium pseudopunctatum* and *R. perssonii*) which originate at leaf axils around the dormant buds. The fact that the flagella of *T. flagellaris* produce such rhizoids or protonemata only at the axils of the scale-like structures corresponding leaves supports the opinion that the burst of rhizoids is genetically controlled and that the different types of rhizoids (micronemata and macronemata in the Mniaceae) found can be used as a taxonomic character.

The protonemata originating from the protonematous rhizoids, the fla-

gella and propagula of the species studied by us show heterotrichous habit which is rather similar to that of the sporeling in *Mnium hornum* (Allsopp & Mitra 1958) and which is in common for most of the moss species (Bopp 1961). In the cultivation only *Trachycystis flagellaris* developed new shoots. However, the yong plants of the *Rhizomnium* grown up from the protonematous rhizoid mat have been frequently observed in the nature. In any case, the present experiments show that the asexual organs and protonematous rhizoids in the Mniaceae have a good growth ability, and, accordingly, it can be assumed that they are a useful mean for short distance dispersal.

Literature

Allsopp, A. & Mitra, G.C. 1958. The morphology of protonema and bud formation in the Bryales. Ann. Bot. **22**: 95-115. Bopp, M. 1961. Morphogenese der Laubmoose. Biol. Rev. **36**: 237-280. Correns, C. 1899. Untersuchungen über die Vermehrung der Laubmoose durch Brutorgane und Stecklinge. Jena. Giles, K.L. & von Maltzahn, K.E. 1967. Interaction of red, far-red, and blue light in cellular regeneration of leaves of *Mnium affine*. Bryologist **70**: 312-315. Koponen, T. 1968. Generic revision of Mniaceae Mitt. (Brophyta). Ann. Bot. Fennici **5**: 117-151. ——. 1971a. A report on *Rhizomnium* (Mniaceae) in Japan. Journ. Hattori Bot. Lab. **34**: 365-390. ——. 1971b. The keys for the Mniaceae in Taiwan. Ann. Bot. Fennici **8** (in print). Lersten, N.R. 1961. A comparative study of generation from isolated gametophytic tissues in *Mnium*. Bryologist **64**: 37-47. Misiura, M. 1964. Regeneracja gametofitu i rozmnazanie wegetatywne *Mnium punctatum* (Schreb.) Hedw. Acta Soc. Bot. Poloniae **33**: 451-459. Watson, E.V. 1967. The structure and life of bryophytes. 2nd ed. 192 pp. London.

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チョウチンゴケ科では *Trachycystis flagellaris* (エゾチョウチンゴケ) のほか，主として *Rhizomnium* 属に無性生殖器官ができることが知られている。本研究は *T. flagellaris* の flagella (鞭枝) と *Rhizomnium* 属の仮根および propagula の発芽パターンを観察するとともにプロトネマの構造についてのべたものである。あわせてこれらの分類学上の意義も論じた。*Trachycystis flagellaris* では flagella の葉腋からプロトネマが生じた。これは成体でも葉腋の部分からのみ macronema (仮根の一種，Koponen 1968 参照) が発生する事実と一致し，この形質は遺伝的に固定した

Trachycystis 属に特徴的なものである。*Rhizomnium minutulum* (コウチワチョウチンゴケ) と *R. striatulum* (スジチョウチンゴケ) の仮根からは，ほとんどすべての部分から，プロトネマがあらたに生じた。*R. tuomikoskii* の propagula では，その先端と，仮根に付着していた部分との両極から，また仮根では，propagula が付着していた部分からのみプロトネマが分化した。*R. tuomikoskii* と同様に propagula をつくる種としては *R. horikawae* が知られているが，両種の propagula の間には形態上，はっきりとした違いが認められる。発達したプロトネマはすべてカウロネマとクロロネマとからなり，異質糸状性をしめした。茎葉体の分化は *T. flagellaris* でのみ観察されている。

○Additional critical taxonomic changes concerning the plants of Okinawa and the Southern Ryukyu Islands (Egbert H. WALKER)

エグバト H. 和嘉： 琉球植物に関する分類学的新知見補遺

The following nomenclatural changes are published here in addition to those published in the Journal of Japanese Botany, **46**: 55–72, March 1971, in anticipation of their need in a coming Flora of Okinawa and the Southern Ryukyus, where it is considered inappropriate to publish such taxonmic and nomenclatural changes.

Ipomoea acuminata (Vahl) Roem. & Schult. forma **albiflora** (Stone) Walker, comb. nov.

Ipomoea indica (Burm.) Merr. forma *albiflora* Stone, in Micronesica **2**: 7. f. 1. 1966.

Ipomoea congesta R. Br. forma *albiflora* (Stone) Walker & Tawada, in Journ. Jap. Bot. **46**: 69. 1971.

This new combination results from the report by S. J. van Ooststroom (ex litt. July 23, 1971) that the name *Ipomoea congesta* R. Br. must be replaced by *I. acuminata* (Vahl) Roem. & Schult. This name is based on *Convolvulus acuminatus* Vahl, Symb. Bot. **3**: 26. 1794, which antedates *I. congesta* R. Br., Prodr. Fl. Nov. Holl. **1**: 485. 1810. Vahl's name became *I. acuminatus* (Vahl) Roem. & Schult. Syst. Veg. **4**: 228. 1819 (not *I. acuminatus* Ruiz & Pav. 1799, an illegitimate name). The trivial forma *albiflora* (Stone) Walker & Tawada, published in March 1971, is therefore here transferred.

Correction: Dr. J. F. Veldkamp of the Rijksherbarium, Leiden, Holland has noted ex litt. that *Zoysia matrella* var. *tenuifolia* (Willd.) Durand & Schinz ex T. Koyama, stat. nov. in Journ. Jap. Bot. **46**: 65. 1971, is a superfluous name. This combination was published in 1928. The following is the correct citation:

Zoysia matrella (L.) Merr. var. **tenuifolia** (Thiele) Sasaki, List Pl. Formosa. 80. 1928.

Basionym: *Z. tenuifolia* Willd ex Thiele, in Linnaea **9**: 309. 1834.—Mauritius.