

## Polyploidy in *Bryum* Collected from the Syowa Station Area, Antarctica

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南極昭和基地周辺から採集された *Bryum* 属の倍数性

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**要旨:** 昭和基地周辺から採集された蘚類 2 種, *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. および *B. amblyodon* C. MUELL. の染色体数について分析した。観察されたほとんどの標本は 2 種とも  $n=20$  を示した。しかしながら, *B. pseudotriquetrum* と同定された一試料に  $n=30$  が見いだされた。これは基本数  $n=10$  の 3 倍体と考えられる。この 3 倍体の配偶体の形態は 2 倍体との間に著しい差は見られなかったが, 葉はより柔らかくしわがより, 葉の細胞はより長く, 葉の先端部は長くとがり, 葉の中肋も長く突出することがある等の特長が認められた。本種の 3 倍体はこれまでソ連で報告されたことがあるが, 倍数性の発生率は両極域など高緯度地方に行くにしたがって増加することが予測される。

**Abstract:** *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. and *B. amblyodon* C. MUELL. collected from the Syowa Station area, Antarctica are karyologically studied with particular regard to the polyploidy. Almost all samples of these two species showed to have  $n=20$ . This may be the diploid level derived from  $x=10$ . In the present study, however, the triploid level ( $n=30$ ) was confirmed in one sample of *B. pseudotriquetrum*, and the morphological features of chromosomes, leaves and cells are compared with those of  $n=20$ . Such polyploidy was hitherto unknown in this species. It is possible to say that the polyploidy increases in incidence with latitude in the polar areas.

### 1. Introduction

Taxonomic studies of *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. and its allied species seem to play an important role in the Antarctic ecosystems. A karyological approach is of great value in analyzing the morphological variation of Antarctic bryaceous mosses. However, only two investigators have so far carried out such studies based on materials from the Syowa Station area, Antarctica (TATUNO, 1963; INOUE, 1976).

Among these karyologically examined mosses, two species of the genus *Bryum*, *B. argenteum* HEDW. and *B. pseudotriquetrum* have been somewhat complicated in taxonomic history. HORIKAWA and ANDO (1961) described two bryaceous mosses from the Syowa Station area, i.e., *B. inconnexum* CARD. endemic to Antarctica and *B.*

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*argenteum* described for the first time in Antarctica. The karyological analysis of the samples was carried out by TATUNO (1963) who concluded that the chromosome number of both species showed  $n=20$ , but the *B. argenteum* was a diploid form ( $n=20$ ) of the species distributed in temperate and tropical zones. However, HORIKAWA and ANDO (1967) suggested that it may be more appropriate to treat it as a variety or even as a separate species from *B. argenteum*. Fifteen years later, the sample of *B. argenteum* with  $n=10$ , a haploid level of  $x=10$ , was collected in the Syowa Station area. As a result, the *B. argenteum* plants with  $n=20$  can be considered a cytotype of *B. inconnexum* (INOUE, 1976; NAKANISHI, 1977). Afterward, OCHI (1979) reported that two bryaceous mosses, *Bryum argenteum* and *B. pseudotriquetrum* from the Syowa Station area and *B. inconnexum* was reduced to a synonym of the latter species. As for the morphological aspects, SEPPELT and KANDA (1986) discussed the potential for variability in Antarctic bryaceous mosses and reconfirmed the identity of many specimens previously determined as *B. argenteum*. In many *B. pseudotriquetrum* plants, the stem leaves showed physical damage near the apex, resulting in the presence of hyaline cells and were consequently easily confused with *B. argenteum*. Moreover, KANDA and OCHI (1986) reported a fruiting plant from the Syowa Station area. The plant was assigned to *B. amblyodon* which was also sometimes confused with *B. pseudotriquetrum*, *B. dichotomum* HEDW., *B. orbiculatifolium* CARD. et BROTH., *B. urbanskyi* BROTH. and *B. pallescens* SCHLEICH.

The existence of such taxonomic problems with Antarctic *B. pseudotriquetrum* underlines the value of undertaking karyological analysis of the species. In the present article, the morphology and chromosome cytology of *B. pseudotriquetrum* are discussed, and new methods of observing the chromosomes in mosses are introduced.

## 2. Materials and Methods

All materials, which were determined either *Bryum pseudotriquetrum* or *B. amblyodon*, were collected from Langhovde, Einstöingen and Rundvågshetta in the vicinity of Syowa Station. The materials, which had been preserved in a freezer at  $-20^{\circ}\text{C}$  for four years from 1983 to 1987, were thawed at room temperature. These were incubated in a large, moist petri dishes at  $15^{\circ}\text{C}$  at alternating 12-hour photoperiodic intervals. The plants were incubated for several weeks and then used for chromosome observations.

### Materials adopted

*Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. Syowa Station area. Langhovde, Yukidori Valley, Coll. H. KANDA, 1. X. 1983 (Nos. 407, 430, 431).

*Bryum amblyodon* C. MUELL. Syowa Station area. Rundvågshetta, Coll. H. KANDA, 11. I. 1983 (No. 274). Einstöingen, Coll. H. KANDA, 18. I. 1983 (No. 423).

Chromosome observations were carried out under the following procedures: Specimens were pretreated with 0.2% colchicine aqueous solution at  $18^{\circ}\text{C}$  for 4 h. Air trapped among tissues sometimes prevents absorption of the solution, so a vacuum pump was used to remove the air. They were fixed with fresh modified Calnoy's

solution (ethanol: chloroform: acetic acid=2: 1: 1) at 5°C for longer than 1 h. They were rinsed with distilled water and macerated in an enzyme mixture (2% cellulase, Onozuka Yakult Co., RS; 0.2% pectolyase, Seishin Co. and 0.6 mol mannitol, pH 5.5) for 15 min at 37°C to soften meristematic tissue. Samples were then rinsed with distilled water again and the meristematic tissue was put on a glass slide and stained with a drop of 2% aceto-orcein for a few minutes. Darkly stained meristematic tissue was destained and mounted with 0.5% aceto-orcein. These were covered with a coverslip and warmed up to 60°C for several seconds, and then squashed. This is the first use of such methods for a karyological study of moss chromosomes.

The voucher specimens used here have been preserved in frozen condition in the National Institute of Polar Research, Tokyo (NIPR).

### 3. Results

The somatic chromosome number  $n=20$  was observed in most of the examined samples of *Bryum pseudotriquetrum*, which were collected from Langhovde (Nos. 407, 431), and *B. amblyodon* from Einstöingen (No. 423) and Rundvågshetta (No. 274) as shown in Table 1. Such a chromosome number is considered to be a diploid level

Table 1. Species, sources and chromosome numbers.

Species	Sources	(Specimen No.)	n
<i>Bryum pseudotriquetrum</i>	Langhovde	(407)	20
	Langhovde	(431)	20
	Langhovde	(430)	30
<i>Bryum amblyodon</i>	Einstöingen	(423)	20
	Rundvågshetta	(274)	20

derived from  $x=10$ . Chromosomes at a late prophase varies in size ranging from 1 to 6  $\mu\text{m}$ , and those at metaphase from 1 to 4  $\mu\text{m}$ . The chromosome complement commonly consists of eight relatively large chromosomes and twelve small ones (Fig. 1: 1).

On the contrary, only a single sample, referable to *B. pseudotriquetrum* from Langhovde (No. 430), showed a peculiar somatic chromosome number,  $n=30$  (Table 1). This was a triploid level derived from  $x=10$ . Chromosomes at a very early metaphase vary in size, ranging from 1 to 5  $\mu\text{m}$ , almost same as in the former samples. The chromosome complement consists of twelve relatively large chromosomes and eighteen small ones, which correspond to one and a half sets in those of the former samples (Fig. 1: 2).

Morphological comparison between these diploid and triploid Antarctic *B. pseudotriquetrum* materials reveals that mosses at the diploid level ( $n=20$ ) are extremely variable but with the following consistent features: the leaves of the specimen No. 431 are larger and long-lanceolate, more rigid and less shrunken; leaf cells are rather shorter; leaf costae sometimes end below the apex; and leaf apex is shortly acuminate (Fig. 2: 1-7). In contrast, the leaves of plants (No. 430) at the triploid level ( $n=30$ ) are more slender and sometimes shrunken; leaf cells are relatively longer; leaf costae are sometimes percurrent; and leaf apex is longly acuminate (Fig. 3: 1-7).

The former bears some superficial resemblance to the *B. pseudotriquetrum* typical

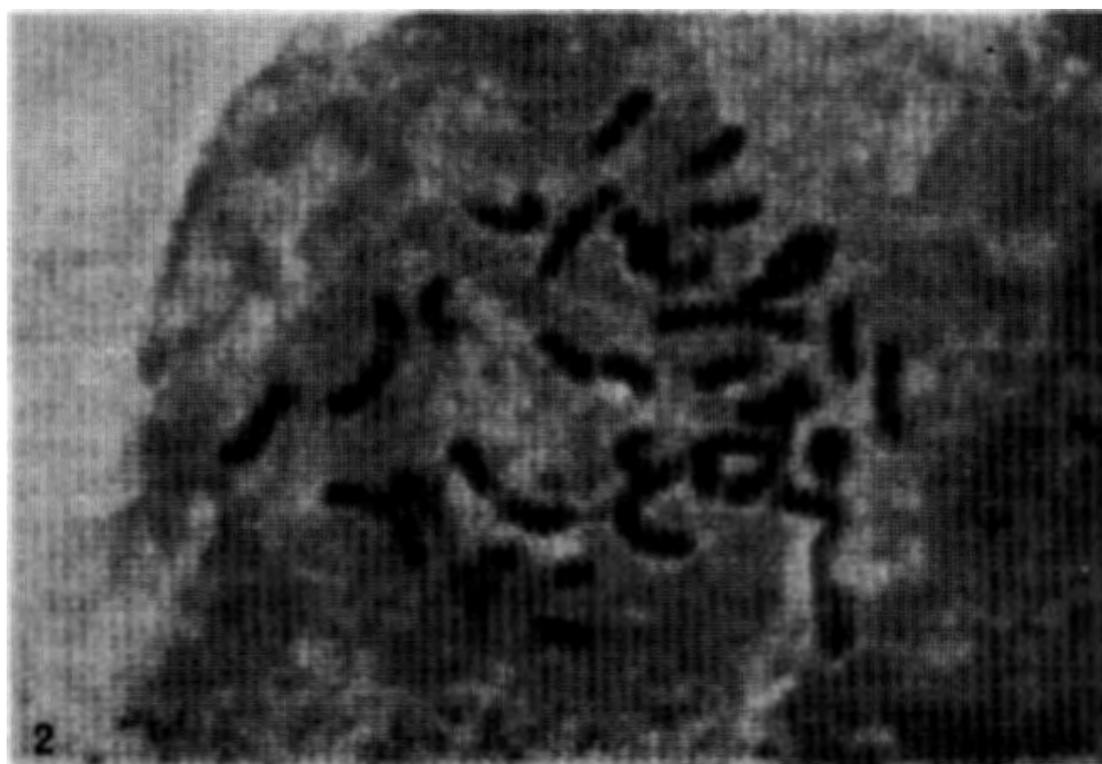
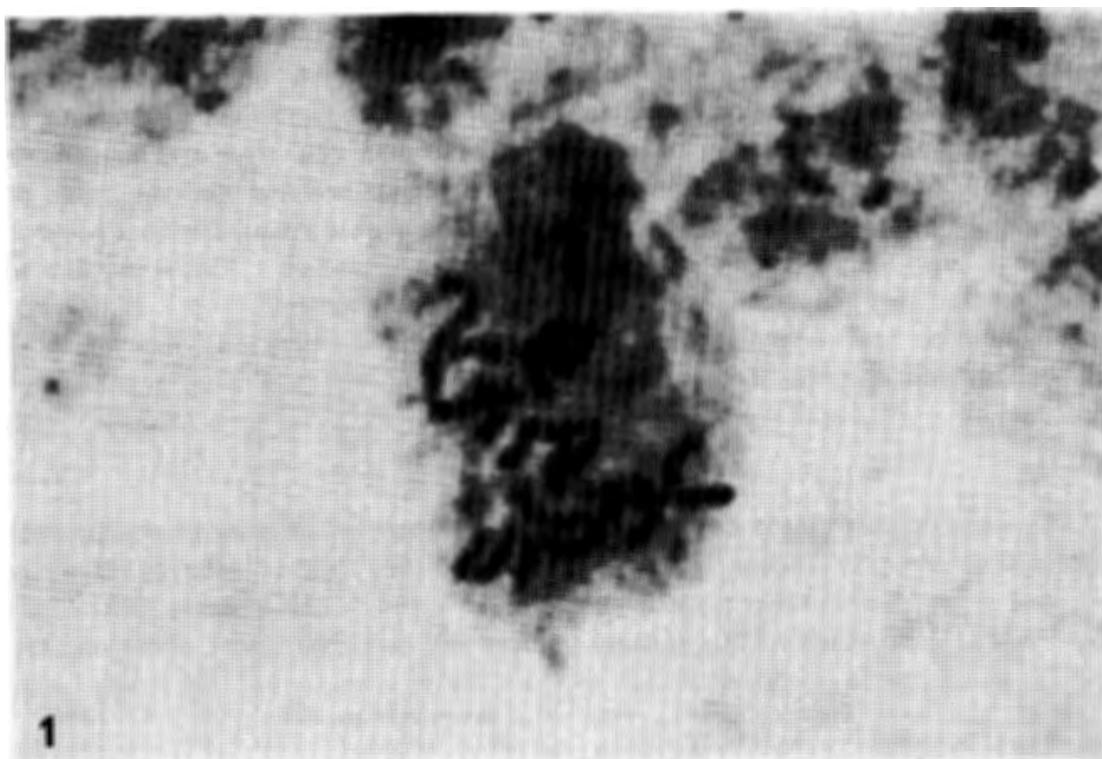


Fig. 1. 1. Chromosomes of *Bryum amblyodon* C. MUELL. showing  $n=20$ .  $\times 4300$ . (No. 274, Rundvågshetta). 2. Chromosomes of *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. showing  $n=30$ .  $\times 4300$ . (No. 430, Langhovde).

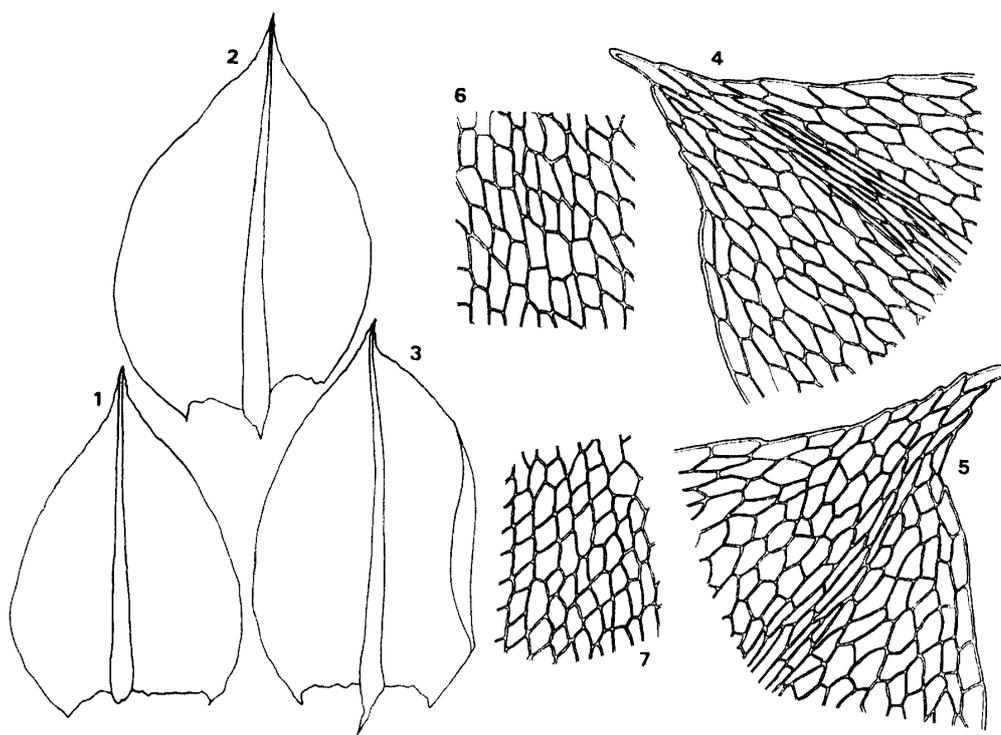


Fig. 2. *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. 1-3. Leaves,  $\times 25$ ; 4, 5. Laminal cells near the leaf apex,  $\times 190$ ; 6, 7. Median laminal cells,  $\times 190$ . (1-7. Drawn from No. 431, Langhovde).

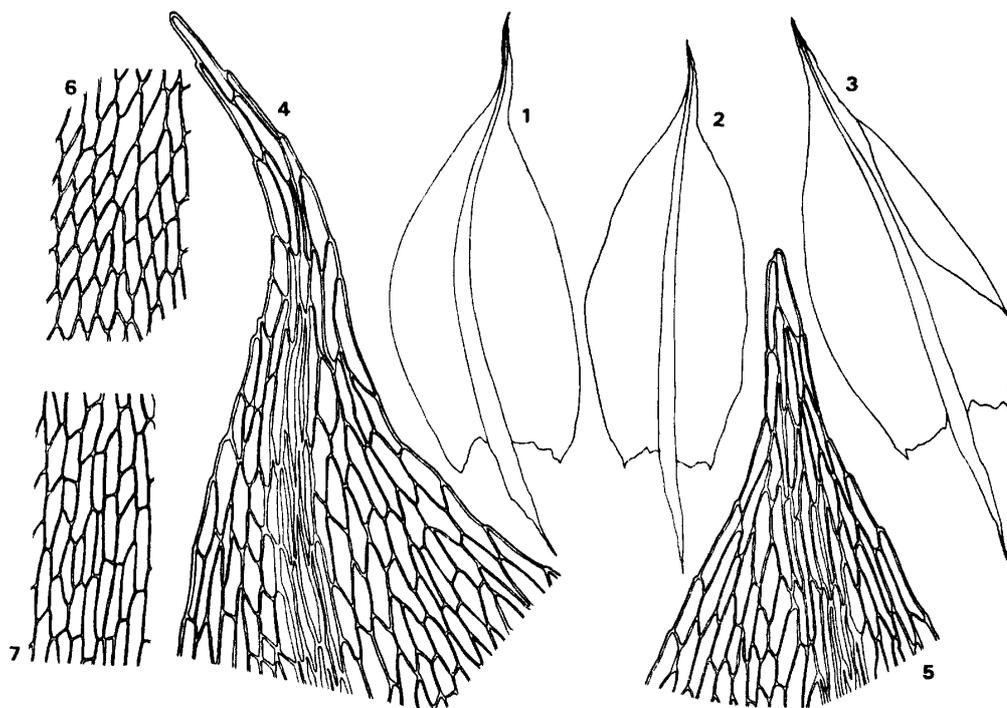


Fig. 3. *Bryum pseudotriquetrum* (HEDW.) GAERTN., MEYER et SCHERB. 1-3. Leaves,  $\times 25$ ; 4, 5. Laminal cells near the leaf apex,  $\times 190$ ; 6, 7. Median laminal cells,  $\times 190$ . (1-7. Drawn from No. 430, Langhovde).

of temperate regions, but the latter resembles the *bimum* type of the species. However, all specimens found in the Syowa Station area are synoicous. On the other hand, two specimens (Nos. 423, 274) of diploid *B. amblyodon* examined are close to the triploid *B. pseudotriquetrum* in vegetative features.

#### 4. Discussion

Karyological studies of Antarctic mosses were first carried out by TATUNO (1963), who counted chromosome numbers of mosses from Syowa Station, East Ongul Island. Two of the three species observed were species of the genus *Bryum*, i.e., both *B. argenteum* and *B. pseudotriquetrum* (as *B. inconnexum*) showed  $n=20$ , as a diploid level derived from  $x=10$ . Following his study, INOUE (1976) investigated mosses from the Syowa Station area, and found euploid series within the genus *Bryum*, namely,  $n=10$  in *B. argenteum* and  $n=20$  in *B. pseudotriquetrum* (as *B. algens* CARD.). NEWTON (1980) also observed  $n=20$  in *B. algens* from Signy Island.

In the present study, a triploid level of  $x=10$  is discovered in one specimen of *B. pseudotriquetrum* from Langhovde, which is the first record for the species. According to FRITSCH (1982), there are polyploid and aneuploid series within *B. pseudotriquetrum*, namely  $n=10$ , 11 and 12 (monoploid level), 20 and 22 (diploid level) and 33 (triploid level). Similarly there are  $n=10$ , 20 and 30 within *B. amblyodon* (as *B. inclinatum* (BRID.) BLAND.). The  $n=33$  in *B. pseudotriquetrum* is reported in Tadzhikistan, USSR by MAMATKULOV (1977) and the  $n=30$  in *B. amblyodon* is reported from Alaska, USA by STEERE (1954). In the global sense, it looks likely that this species exhibits increasing polyploidization accompanied by the expansion of the distribution towards higher latitudes. The monoploid level of  $x=10$  is commonly known in temperate and/or tropical zones, while higher ploidy levels are distributed at rather higher latitudes. Considering such a trend, it is a remarkable fact that both diploid (No. 431) and triploid (No. 430) plants were discovered in the same stream area of Langhovde, Antarctica. It is presumed that the phenomenon of habitat segregation between ploidy levels plays a role in this species under rather severe Antarctic environmental conditions. However, the specimens so far examined are insufficient to discuss the relationship between the morphology and the chromosome cytology in further detail.

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