

ON THE EXPANSION OF *ATRICHUM FLAVISETUM* IN MOSCOW PROVINCE:
HOW DOES IT DIVIDE HABITATS WITH *A. UNDULATUM*

О РАССЕЛЕНИИ *ATRICHUM FLAVISETUM* В МОСКОВСКОЙ ОБЛАСТИ:
КАК ОН ДЕЛИТ МЕСТООБИТАНИЯ С *A. UNDULATUM*

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Abstract

Atrichum flavisetum was a rare species in 19th and 20th centuries in Moscow Province, where it grew on soil on eroded slopes of exceptionally deep ravines. However, since the first decade of 21st century it started to spread in old-growth forests, inhabiting soil banks under upturned roots of fallen trees. Observations in two recent decades in the reserved forest area at the Zvenigorod Biological Station of Moscow State University, near Moscow documented its invasion and tendency of partial substitution of the most common indigenous species *Atrichum undulatum*, which occurred in such habitats in 19th and 20th centuries. However, it is likely that in a relatively flat places *A. flavisetum* is unable to compete with *A. undulatum*. A comparison of morphology of these two species revealed the differences in one of the largely neglected structures, the rhizoidal ropes, that has been sometimes interpreted as underground rhizome, which is actually absent in *Atrichum*. In both species the rhizoidal ropes are highly hygroscopic and capable for vigorous movements, making effort possible for moving a weight three times exceeding the weight of whole plant itself. The ‘rhizoidal rope force’ is studied and shown to be stronger in *A. undulatum* than in *A. flavisetum*. The comparison of the rhizoidal rope structure in these two species suggests stronger structural differentiation of rhizoids in *A. undulatum*. Such differentiation probably makes the hygroscopic movements stronger and favors survival of *A. undulatum* on the more dense soil where *A. flavisetum* doesn’t grow, and it results in a division of preferable habitats between these two *Atrichum* species.

Резюме

Atrichum flavisetum в XIX и XX веках был редким видом в Московской области, где рос на оползающих склонах наиболее глубоких оврагов. Однако в первом десятилетии XXI века он начал расселяться в старовозрастных лесах на обнаженной почве выворотов в основании упавших деревьев. Наблюдения на заповедном участке леса на Звенигородской биостанции МГУ под Москвой за последние два десятилетия документируют его появление, поступательное расселение и частичное вытеснение им широко распространенного в подобных местообитаниях *Atrichum undulatum*. Однако на б. м. плоских участках обнаженной почвы в лесах *Atrichum flavisetum* вряд ли будет способен конкурировать с *A. undulatum*. Среди слабо изученных признаков *Atrichum* особое внимание уделено тязам ризоидов, часто неверно интерпретируемым как “подземные корневища”, которые у *Atrichum* отсутствуют. Такие тязи, особенно хорошо развитые у *A. undulatum*, представляют собой ризоиды, расположенные по спирали вокруг каулономы; они обладают высокой гигроскопичностью и силой, достаточной для того, чтобы двигать объекты, превышающие вес самого растения в 3 раза. Большая ‘сила ризоидных тязей’ у *A. undulatum*, чем у *A. flavisetum*, в определенной степени связана с их более сложным строением, в частности, с более сильным изменением ризоидов в процессе формирования тязя, что, в свою очередь, может способствовать его произрастанию на более плотной почве, на которой *A. flavisetum* не растет. Таким образом, у этих видов *Atrichum* складывается вполне определенная дифференциация по типам местообитаний.

KEYWORDS: species competition, *Atrichum undulatum*, new habitats, rhizoids, biomechanics

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INTRODUCTION

Moss dispersal by spores makes them potentially a quite dynamic component of ecosystems. However, moss species have generally wider distribution than species of vascular plants, and it hides long dispersal events and makes the number of invasive moss species much fewer than those in vascular plants. Attribution of moss species in certain area to invasive is in many cases equivocal (cf. Essl *et al.*, 2014, 2015; Patiño & Vanderpoorten, 2015).

Rapid expansions of the best-known examples of invasive species, *Campylopus introflexus* (Hedw.) Brid. and *Orthodontium lineare* Schwägr., illustrate that invasions may proceed in natural habitats, contrasting to vascular plants where recently introduced species occur mostly in ruderal and otherwise strongly disturbed environments. One of examples of rapid spreading in the Central European Russia is provided by *Schistostega pennata* (Hedw.) F. Weber & D. Mohr, which became a common species after changes in forest management. The forest usage for recreation led to more numerous tree windfalls in partly protected forests, with more numerous soil banks under upturned roots of fallen trees (Ignatov & Ignatova, 2001), creating habitats suitable for *Schistostega*. The present paper addresses to another species with similar ecology and similar way of expansion, *Atrichum flaviseum* Mitt.

Atrichum flaviseum is a member of *A. undulatum* (Hedw.) P. Beauv. complex, which has a complicated taxonomy. For a long time *A. flaviseum* was treated as merely an intraspecific entity, *A. undulatum* var. *gracilisetum* Besch. (Nyholm, 1961). A detailed study of *A. undulatum* has been conducted recently in the North America (Perley & Jesson, 2015), while in Eurasia it is still awaiting a study with molecular markers. Regardless the uncertainty with proper names for all Eurasian taxa of autoicous *Atrichum* species, the Central European Russian plants of autoicous *Atrichum* include two distinct species easily separated in the field by 2–4(–7) sporophytes per plant in *A. flavisetum* vs. 1(2) in *A. undulatum* s.str. (Fig. 1).

Atrichum undulatum s.str. is one of the most widespread species in Moscow Province (hereafter accepted as including territory belonging to Moscow City, the administratively separate unit from Moscow Province), growing in forests on disturbed soil, commonly along trails, both on flat places and on vertical banks, and also on soil dug out by moles and various rodents (e.g. *Clethrionomys*), on soil covering roots of fallen trees, on landslides, and other habitats suitable for colonization. *Atrichum flaviseum* was known in Moscow Province in very few localities in 19th century (Zickendrath, 1900) and most of 20th century, occurring in deep ravines, in habitats where it was often associated with a rare fern, *Polystichum braunii* (Spenn) Fee, so Ignatov & Ignatova (1990) published the map of co-localization of these two species.

Altogether, 12 localities of *A. flaviseum* were recorded in Moscow Province by 1990, thus the species was a subject of protection, and it was redlisted for the territory of Moscow, where its populations in ravines were de-

clining. Contrary to this, in more eastern parts of Russia, in Middle Urals and Siberia (e.g. Altai), *A. flaviseum* is a forest species, growing on soil covering roots of fallen trees, while *A. undulatum* occurs mostly on soil along trails.

Changes in *Atrichum flavisetum* ecology were noticed first in 2005, at the Zvenigorod Biological Station, the instant area for summer classes for students of the Faculty of Biology of the Moscow State University. *Atrichum undulatum* was and is there a very common species, whereas *A. flaviseum* was not recorded until 2005, despite of extensive moss studies conducted there in the 1990s and 2000–2001, with a none record of this species. After a break of observation in years 2001–2004, in 2005 *A. flaviseum* was found in several localities at roots of fallen trees (Fig. 1), and in 2006 it was mapped, showing its presence on 40 out of 46 studied ‘tree-fall upturns’, prevailing *A. undulatum* in abundance in about one third of them (Ignatova & Ignatov, 2011).

Later, in 2012, 2018, and 2021–2024, observations with counts were conducted in order to trace a possible further expansion of *A. flavisetum*, compare its habitats with those of *A. undulatum*, and seek characters which potentially may explain some difference in their ecology.

METHODS AND MATERIALS

Study area

Field observations were conducted at the Zvenigorod Biological Station of the Moscow University (ca. 55.7°N, 36.6°E). Pine and spruce forests in its territory were mostly planted in 19th and early 20th centuries. Since 1950s the forest became protected by prohibiting any clear-cuttings. The forest became older, with more frequent tree falls and subsequent replacing the conifers by *Betula pendula*, *B. pubescens*, and broad-leaved trees (mostly *Tilia cordata*, *Acer platanoides*, and *Quercus robur*). However, old growth spruce forests still are widespread in this territory, and the state of a conserved area causes the presence of numerous fallen trees.

Since 2006, observations were conducted during summer classes, and in some years a small student projects were performed, aiming to monitoring the spreading of *A. flavisetum* and its competing with aboriginal in the biostation *A. undulatum*.

Supplementary observations of the distribution of *Atrichum flaviseum* were done in places potentially suitable for it: (1) in other habitat types at the Zvenigorod Biological Station; (2) in other forests near Moscow (ca. 55.7–55.8N, 37.3–37.4E).

Field observations included counts of the relative abundance of species and the number of sporophytes. Soil acidity has been measured for several ‘typical’ habitats of each species.

Rhizoidal rope study

Unusual structure of *Atrichum undulatum* s.l. rhizoids that swirl around, forming complex ropes, has been very clearly illustrated by Schimper (1860). Ruhland



Fig. 1. Habitats and habits of *Atrichum flavisetum* and *A. undulatum*. A: average look of 7–10 years old upturn of a fallen tree; B: *A. flavisetum* on upper ledge; C: *A. undulatum* on upper ledge; D: *A. flavisetum* on subvertical soil bank; E: *A. flavisetum* (F) and *A. undulatum* (U) from mixed populations; note that U plants are larger than F ones.

(1924) reproduced Schimper' pictures in the introduction to the Brotherus' volume in the famous 'Die natürlichen Pflanzenfamilien' of Adolf Engler, but erroneously referred Schimper' drawings to *Polytrichum jupinerinum*. The latter species also belongs to Polytrichaceae and has a principally the same structure of underground organs, but to a much lesser extent, so it is sometimes not easy to find such rhizoidal ropes. Probably this 'misaddressing' caused the fact that rhizoidal structures of *Atrichum* were not in the focus of the studies of bryologists, as far as we know. Duckett *et al.*' (2004) study of rhizoidal morphology in the *in vitro* culture included one of *Atrichum* species, but the rhizoidal structure has been shown for *Polytrichum commune*, where the rhizoidal ropes are formed by a relatively few rhizoids compared to *Atrichum undulatum*.

Coming to the study of *Atrichum flavisetum* and *A. undulatum* putative competition, we considered, among others, their underground structures and the rhizoidal ropes specifically. They were studied by their morphology by a standard method of light microscopy. SEM studies were conducted under SEM Thermoscientific Quattro S equipped with a field emission gun. Material cut by razor blade has been glued on the carbon tape upon aluminum stubs and observed without gold or platinum coating. Observations were conducted at the accelerated voltage 15 kV, at environmental SEM (ESEM) mode, at the air pressure ca. 500 Pa, under 15 kV, using SE mode.

To prepare anatomical sections, the stem bases with rhizoidal ropes were fixed in a 2.5% glutaraldehyde solution for 7 days. After washing, the material was dehydrated in an alcohol series (20%, 40%, 60%, 80% and 96% alcohol), alcohol-acetone mixture (1:1), and acetone for 1 hour in each solution, soaked in an acetone-resin mixture series (3:1, 1:1, 1:3) for 12, 24 and 3 hours respectively, and embedded in epon-araldite resin as recommended by the manufacturer. The resin was polymerized at 60°C for 24 hours. Serial transverse sections were cut 2 µm and 1 µm thick with glass knives, placed on glass slides without mounting medium and studied with at 40x/0.9 objective lens for autofluorescence, without staining under LOMO fluorescent microscope BLM-L with ultraviolet filter 330-385 nm (with mercury lamp), equipped by digital camera MC-20.

During material preparation for microscopic studies, a high hygroscopicity of rhizoidal ropes was noticed and their movements then became an additional part of the study. These movements were recorded on video in 2023. In 2024 we addressed specifically to 'traction force' of rhizoidal ropes of two species. For the latter, *Atrichum undulatum* (*A. undulatum* 1) and *A. flavisetum* were collected in the same plot, 55°42'04.58"N 36°43'28.06"E. Original plan supposed to compare plants growing in a fully identical environmental condition. The habitat was the upper part of an upturn of ca. 2 m high, i.e. the extensive soil wall on roots of a big fallen spruce tree in shady spruce forest. A similar but smaller upturn is shown in Fig. 1A.

The results obtained from the mentioned plot found too small difference between two species, and therefore additional samples were added for *A. undulatum* from denser soil along a trail nearby (*A. undulatum* 2) and in Moscow, along a trail in oak wood (*A. undulatum* 3). Only *A. undulatum* was studied from such habitats, as *A. flavisetum* has never been observed growing in flat places (both in the study area and in the Central European Russia as a whole).

Collected material was washed as much as possible with water and then separated into individual plants, each possessing one rhizoidal rope. The length of their aerial part, rhizoidal rope length and the width at 1–2 mm from the stem base were measured.

To determine the 'traction force' of the rhizoidal rope, graph paper with glued on its back side double-sided tape was used. A number of 'weights' with the known weight (1 mm² = 0.153 mg), were prepared ahead of measurements. Their weights did not change during at least eight hours, as could be suspected considering tape drying. Tests used weights from 5 mm² to 120 mm², and most rhizoidal ropes were able to carry 'weights' up to 40 mm² (i.e. 6.1 mg).

After the 'weight' was attached, the mosses were placed in a Petri dish, under which there was graph paper for accurate assessment of the movements of the rhizoidal ropes, after which the movement intensity (MI) during wetting and drying cycles was assessed with a three-point scale: 1 – slow linear movements; 2 – some linear and a small rotational movements; 3 – intense rotational and linear movements. The stem of the tested moss was fixed down with a heavy object to exclude any movements. Evaluation of a 'rhizoidal rope force' (RRF) was determined as a product of maximal weight and movement intensity (MI).

Stereomicroscope Olympus SZX16 with a built-in "Infinity.2.2" camera with a resolution of 2080x1536 were used for data recording.

RESULTS

Atrichum species abundance within the area

The relative abundance of *Atrichum undulatum* and *A. flavisetum* on soil at upturned roots of fallen trees in 2006, 2018 and 2021–2023 is shown in Table 1. Despite of only little changes, in overall abundance of *A. flavisetum* is slightly growing in some places: e.g. in a ravine where only *A. undulatum* occurred in 2018, in later years, 2023–2024, *A. flavisetum* was recorded in many places.

The population density has been counted for three

Table 1. Number (N) of soil banks under upturned roots of fallen trees at the Zvenigorod Biological Station where *Atrichum flavisetum* (F) or *A. undulatum* (U) or both (F+U) were recorded from 2006 to 2023.

Year	N	F+U,%	F,%	U,%
2006	46	80.5	6.5	13.0
2018	53	71.7	7.5	20.8
2021	48	75.0	16.7	8.3
2022	60	81.7	6.7	11.6
2023	26	61.5	7.7	30.8

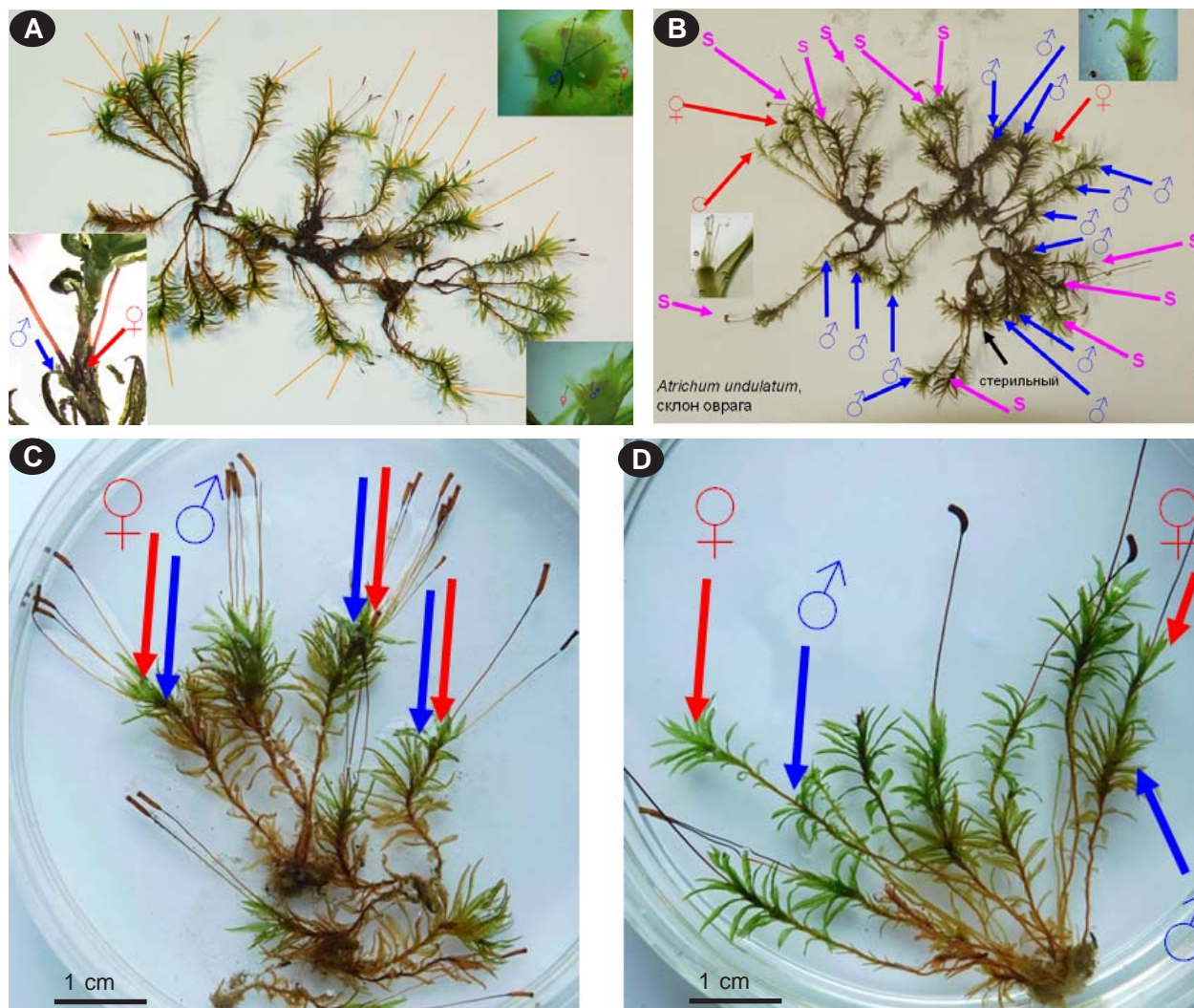


Fig. 2. Distribution of male (blue), female (red) and paroicous (yellow) inflorescences in *Atrichum flavisetum* (A, C) and *A. undulatum* (B, D); S – sterile plants (pink).

plots 10 × 5 cm each for both species in early June, the time when capsules are well developed. Number of shoots without capsules (S) / shoots with capsules (C) was found to be as follow:

A. flavisetum: 194 S / 203 C; 335 S / 364 C; 435S / 430 C

A. undulatum: 521 S / 229 C; 428 S / 105 C; 312 S / 186 C

The similar results for the same size plots were obtained in late July, when capsules were partly fallen off, provided the similar counts by habitats from 5 × 3 cm plots (Table 2). In addition, the angle of slope inclination and soil pH were measured for each plot. Our data found no substantial difference in soil pH and number of shoots per plot. At the same time, it is clear from these observations that *A. flavisetum* almost never grows in flat places.

Table 2. Number of shoots of *Atrichum undulatum* (U) and *A. flavisetum* (F) per plot of 5 × 3 cm, the angle of slope inclination, and pH of soil under the tuft.

#	Habitat	U, n	U, incl.	U, pH	F, n	F, incl.	F, pH
15	upturn (U above F)	86	50	4,95	102	67	4,8
16	upturn	–	33	5,01	–	22	5,04
17	upturn, old	82	60	4,81	0	–	–
18	upturn, mix of U+F	115	75	4,84	146	75	5,29
12	ravine	173	84	5,37	99	45	5,15
13	ravine	72	62	5,48	125	63	5,25
11	soil bank along trail	76	54	4,64	74	50	4,66
19	soil bank along trail	109	34	4,5	0	–	–
20	trail side, flat	99	10	4,7	0	–	–
22	trail side, hummock	84	23	4,58	0	–	–
23	trail side, flat	147	14	4,82	0	–	–

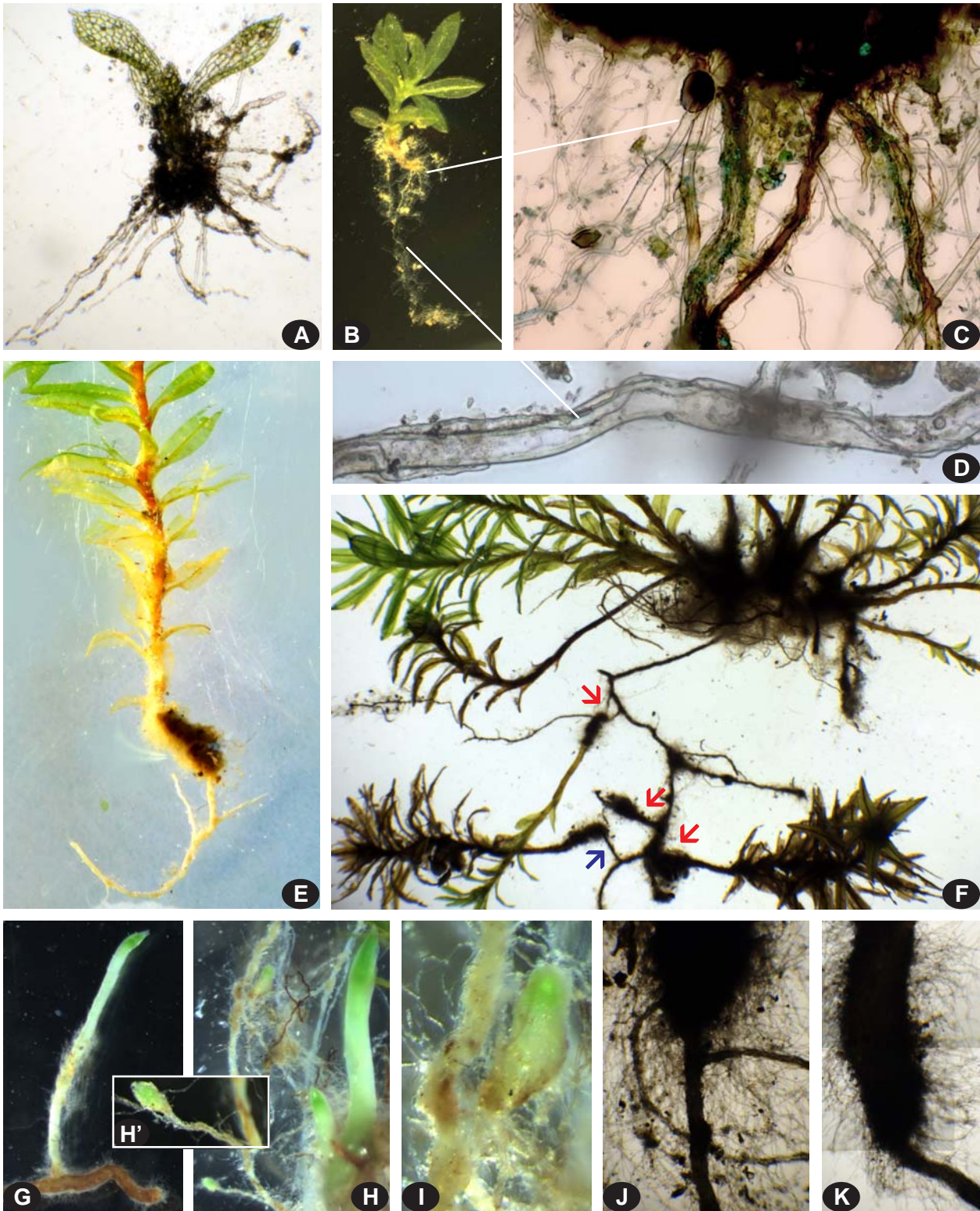


Fig. 3. Rhizoidal ropes of *Atrichum undulatum*: from young (A–E) to mature (F, J, K) plants; G–I: innovations on the underground rhizoidal ropes.

Rhizoidal rope structure

Rhizoids occur in *Atrichum* plants as a cluster of whitish filaments already at a very early stage of development, when the stem has only two leaves, which are still ecostate (Fig. 3A). The species identity is not possi-

ble to decide at this stage. Originally rhizoids look all the same, but since the stage of (5)10 leaves (Fig. 3B), one among them looks unequivocally thicker (Fig. 3C, 3D). As subsequently such thick filaments are found with bud initials which are further developing in new shoots,

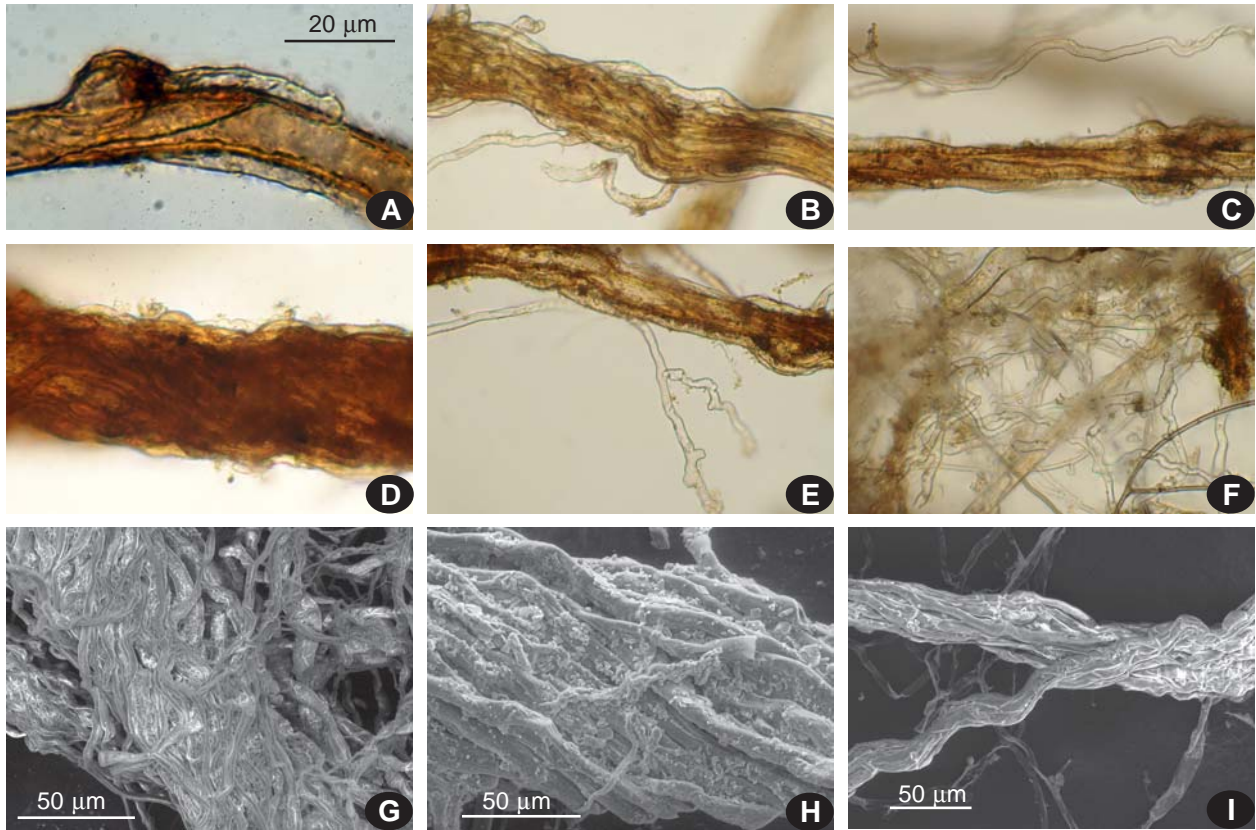


Fig. 4. Rhizoidal ropes of *Atrichum undulatum* (A–H) and *A. flavisetum* showing variation, and contorted rhizoids (E, F). A–F: under light microscope, G–I: under SEM: note a greater pitch angle of the spirals in the former species.

we call them ‘caulonemata’, following the terminology used by Duckett *et al.* (2004). Smaller rhizoids that branch off this caulonemata start to swirl around it already at the stage of a ten-leaved plant (Fig. 3D), and at the stage of 1 cm tall plant (Fig. 3E) the rhizoidal rope looks well-formed, obviously with more than one layer of thinner rhizoids upon the caulonemata.

Rope thickening proceeds gradually, and finally they reach to about 100 µm wide. Such ropes form the underground net connecting a number of shoots (Fig. 3F). Despite of the solid cover of small rhizoids densely covering the core ‘strand’, the buds originate on caulonemata, immediately on it (Fig. 3G,I,J) or, occasionally, on thin branches, also caulonemata (Fig. 3H). The joints of rhizoidal ropes with the stem are usually well seen (Fig. 3F (arrows), K, L).

The variation of rhizoidal ropes within almost each specimen is great. In different parts of rhizoidal mass, there are thick caulonemata with only a few rhizoids which start their creeping growth upon the caulonemata surface (Fig. 4A), and quite thin rhizoid ropes with rhizoids apparently embracing rather thin caulonemata (Fig. 4B–C).

Rhizoids are usually spirally twisted around caulonemata (4D), but the pitch angle of these spirals varies from ca. 70° to approaching zero. No correspondence of this pitch angle to the rope width was observed. Even within one rope the spiral appearance varies greatly.

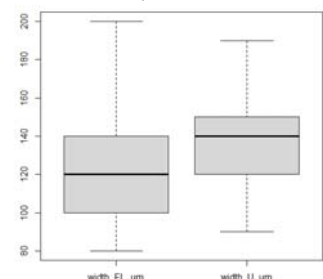
However, we noticed, albeit by an only preliminary observation, the greater angle in *A. undulatum* than in *A. flavisetum* (Fig. 4G–I), while in the latter the rhizoids on caulonema are often almost straight.

Thin rhizoids spreading from the rhizoidal ropes usually end in a strongly flexuose, contorted and irregularly thickened parts (Fig. 4E, F).

It is more difficult to make transverse sections of rhizoidal ropes than, e.g. of leaves, due to their rather soft structure, especially in *A. flavisetum*. The anatomy of them vary from one section to another, hampering the application of statistical or otherwise quantitative characteristics (but a short comments are given below in the Discussion section).

The results of measure of the rhizoidal rope width near the base of the stem is shown in Fig. 5. It shows that in *A. undulatum* rhizoidal ropes are thicker, though strong overlap makes the significance of this difference low (p -value = 0.1, by Kruskal & Wallace test).

Fig. 5. Rhizoidal rope width, at ca. 1 mm from stem base of *Atrichum undulatum* ($n = 50$) and *A. flavisetum* ($n = 50$).



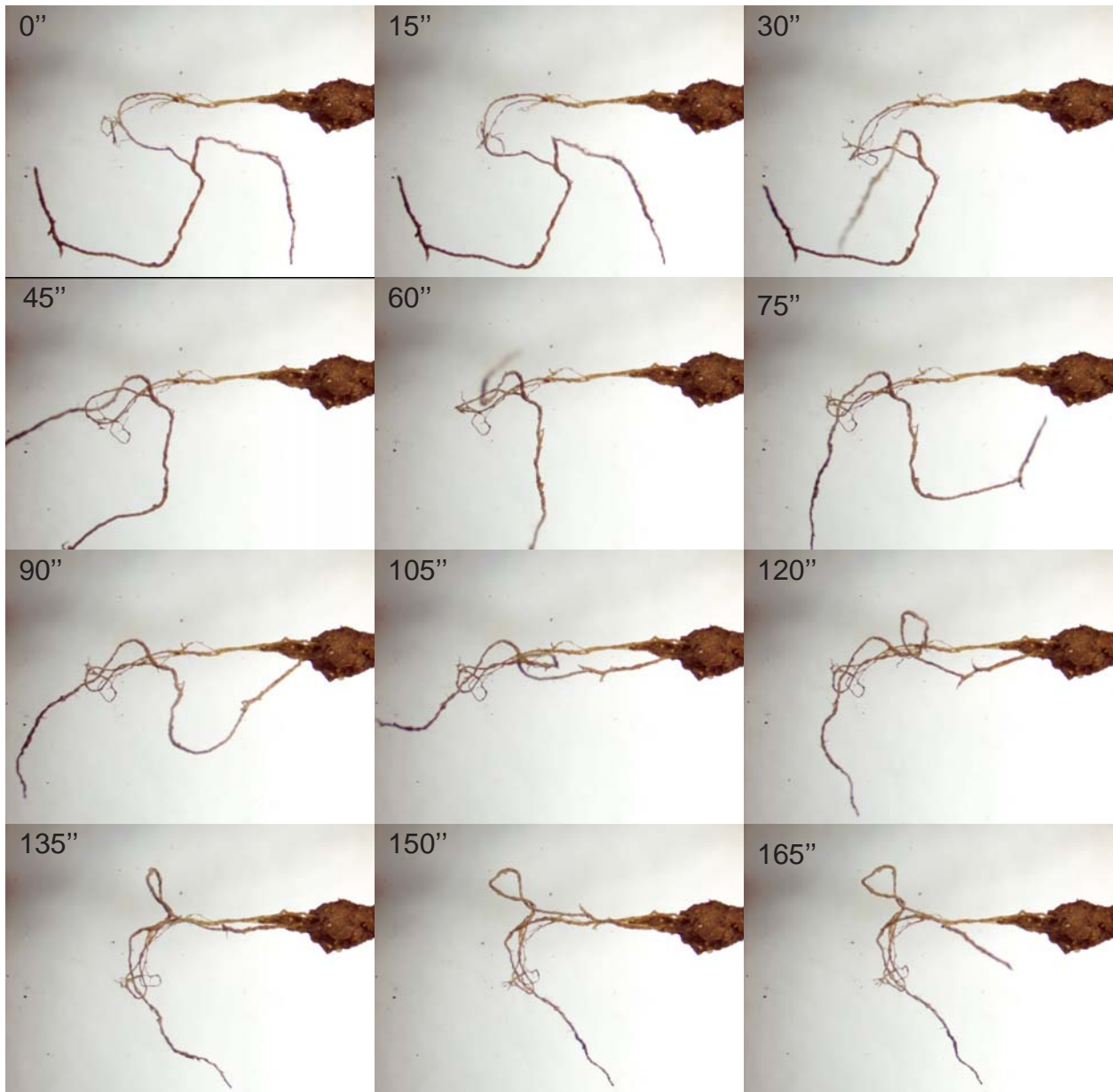


Fig. 6. The real-time movements of *Atrichum undulatum* rhizoidal rope during drying within 165 seconds. Pictures taken from video available in fast (10x) variant at `u_drying_short.mp4` (Supplementary Materials 2).

Rhizoidal rope movements

Based on a general observation, the rhizoidal ropes of *A. undulatum* perform more intensive and faster hygroscopic movements than rhizoidal ropes of *A. flavisetum*. Such movements appeared both after wetting a dry rhizoidal rope (by a drop, or by a sinking in the water of the whole plant), and during the plant drying after being moistened that hygroscopic movements are sometimes quite conspicuous. One of such series that lasted three minutes is shown in Fig. 6. The stem has been fixed by pressing of a weight, the video was recorded by the digital camera of the stereomicroscope. This and other similar videos are available in the Supplementary Materials 2 (https://kmkjournals.com/journals/Arctoa/Arctoa_33_226_238_video).

Supposing that the major question of the present study about the possible declining of *A. undulatum* population as a result of *A. flavisetum* invasion may relate to the underground organs of these species, we attempted to evaluate the 'rhizoidal rope energy' not only visually. The measuring of maximal weight that rhizoidal rope can pull during the drying, or push during the wetting, or turn around in the course of spiral twisting, or move in a its own specific way, was provided.

The preliminary tests found that some movements are fast and strong, whereas others are hardly noticeable, so this character, the Moving intensity (as explained in Materials and methods) was estimated for each event. Fig. 7 illustrates such variants.

The 'rhizoidal rope force' (RRF, cf. Material and

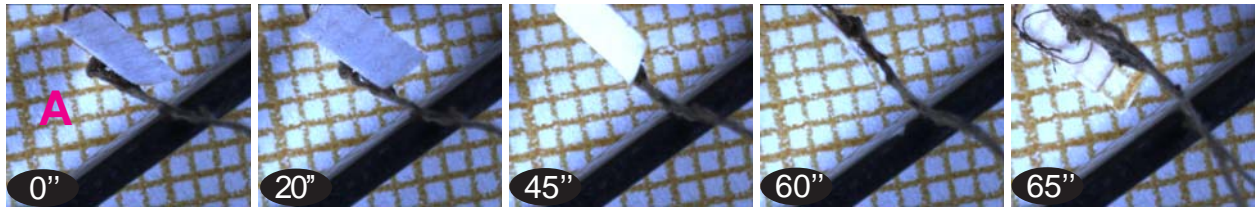


Fig. 7. Examples of hygroscopic movements of rhizoidal ropes, moving weights. Real time is shown in seconds.

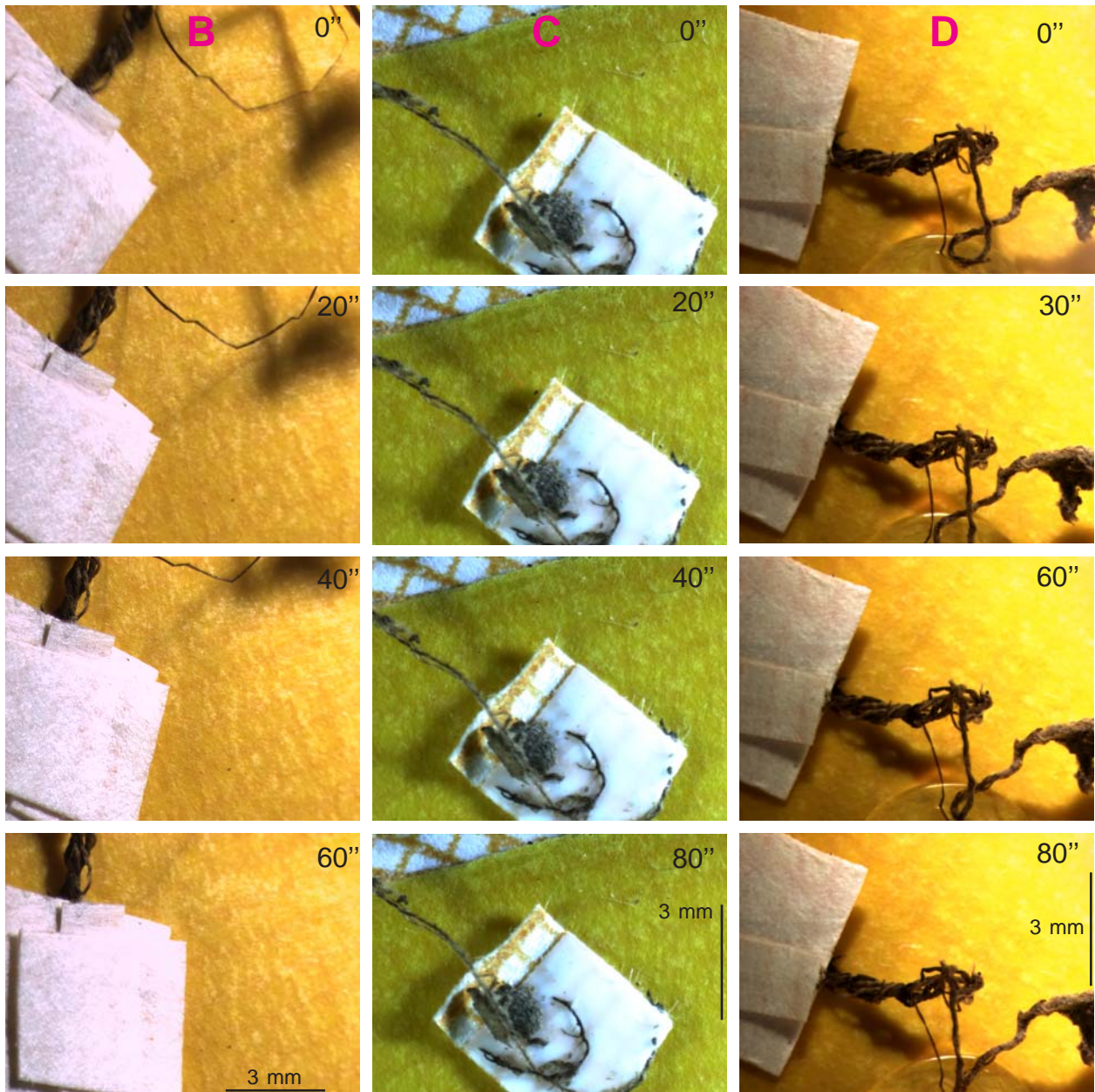
A: *Atrichum flavisetum*, weight=0.75 mg, fast moving and turning (MI=3) during drying; f-1-34_10mm_move_dry - 4x.mp4

B: *Atrichum undulatum*, weight=10 mg, moderate moving (MI=2) during wetting; u-2-5-66mm-move-wetting2.mp4

C: *Atrichum undulatum*, weight=3 mg, slow moving (MI=1) during drying; u-1-50_20+mm_move_dry - 4x.mp4

D: *Atrichum undulatum*, weight=7 mg, slow moving (MI=1) during drying; u-2-5-46mm-move-wetting - 4x.mp4

Video available at Supplementary Materials 2.



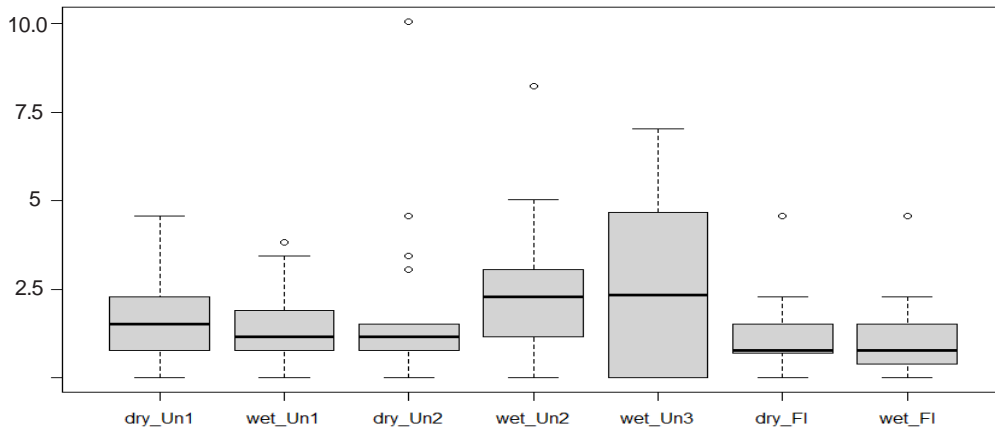


Fig. 8. Rhizoidal rope force (in mg of moved weights) of hygroscopic movements of *Atrichum undulatum* and *A. flavisetum*, for drying and wetting tests. Data are given in Supplementary Materials 1.

methods) for wetting and drying was found in originally studied *A. flavisetum* and *A. undulatum* 1 to be fairly similar. This is not necessarily the rule for all samples, and, for example, for *A. undulatum* 2 the wetting and drying RRF appeared to be quite asymmetric in this respect. For calculation total results, we used RRF from wetting and drying tests.

Altogether the tests were conducted for 21 plants of *A. flavisetum*, *A. undulatum* 1, *A. undulatum* 2, and 50 for *A. undulatum* 3. Each plant has been tested for drying and for wetting, thus altogether 226 tests were done. The strategy to find the maximal weight for an individual sample depends on preliminary observation of its moving intensity with small (ca. 1 mg) weight.

Our first comparison of *A. flavisetum* and *A. undulatum* 1 (from the upturn) found RRF in the former species to be lower, but statistically insignificant (Table 4 in SM, Fig. 8). Subsequent tests with *A. undulatum* 2 and 3 demonstrated that the *A. undulatum* RRF is often greater. The difference between *A. flavisetum* and *A. undulatum* 3 from the soil in broad-leaved forest near trail differ statistically significant (p -value = 0.006802). However, the difference between all tested specimens of *A. undulatum* and *A. flavisetum* is not significant (Fig. 8).

It is interesting that the difference between *A. undulatum* 1 and 2, from the same area, but growing on a rather soft and more dense substrate, is significant, albeit low (p -value = 0.041).

The maximal weight moved by *A. undulatum* 1 (from the upturn) was 4.58 mg, and by *A. flavisetum*, also from the upturn, 3.81 mg. Plants of *A. undulatum* 2 from denser soil along a trail moved 10.07 mg, which is more than twice greater its own weight, 4.8 mg, including stem with leaves and underground portion. An additional search, when the work was largely accomplished, revealed a sample which moved the weight 15.25 mg (100 mm²). Plants from the autumn collection from dense soil, *A. undulatum* 3, were able to move weight over 8 mg more often than samples of *A. undulatum* 1.

Thus, having no significant difference in mean rhizoidal rope force, *A. undulatum* has the ability to move greater weights, especially when plants have grown on dense soil.

DISCUSSION

Observations within 20 years after the first record of the species at the Zvenigorod Biological Station show a quite slow expansion of *A. flavisetum* in the forest where the numerous tree windfalls occur and continue to reappear. The main difference from the original observations in 2005–2006 is that now *A. flavisetum* occurs more often on the upper, more or less horizontal part of upturns. This can be explained by the upturn eroding, so their upper parts become softer, resembling the subvertical faces of an upturn of just fallen trees. The more numerous findings of *A. flavisetum* on the eroding ravine slopes nearby is another evidence of the general species spreading.

The flat sites with dense soil are however not suitable for *A. flavisetum*: only once few plants of this species were noticed on a molehill, but they did not persist there for the next year.

What are the features which favor *A. undulatum* to grow in flat places where *A. flavisetum* fails? To find traits which might correlate with this, we undertook some more studies of the rhizoidal rope structure. We are presenting them now, within the discussion, as they are only preliminary, based on a limited number of samples used for comparative studies of 'rhizoidal rope force', in order to point possible directions of further search the differences between these two species, and maybe other species, also in other regions.

The transverse sections made by razor blade and studied under SEM, without coating, at 'environmental mode' of ca. 500 Pa air pressure, show stronger cell walls thickenings in *A. undulatum* compared with thinner, more smooth cell walls in *A. flavisetum* (Fig. 9). These images show a strong aggregation of rhizoids around caulone-mata, thus, at first glance, one may assume that the images show the tissue, not the tightly packed rhizoids (both in *A. undulatum*, Fig. 9G, and *A. flavisetum*, Fig. 9I). The less tightly packed structure is better seen in oblique sections in *A. flavisetum* (Fig. 9H). The rhizoid cell walls in the latter species are smooth (Fig. 9C, F, H), whereas in *A. undulatum* they look as covered by additional material, making cell walls rough (Fig. 9D, G).

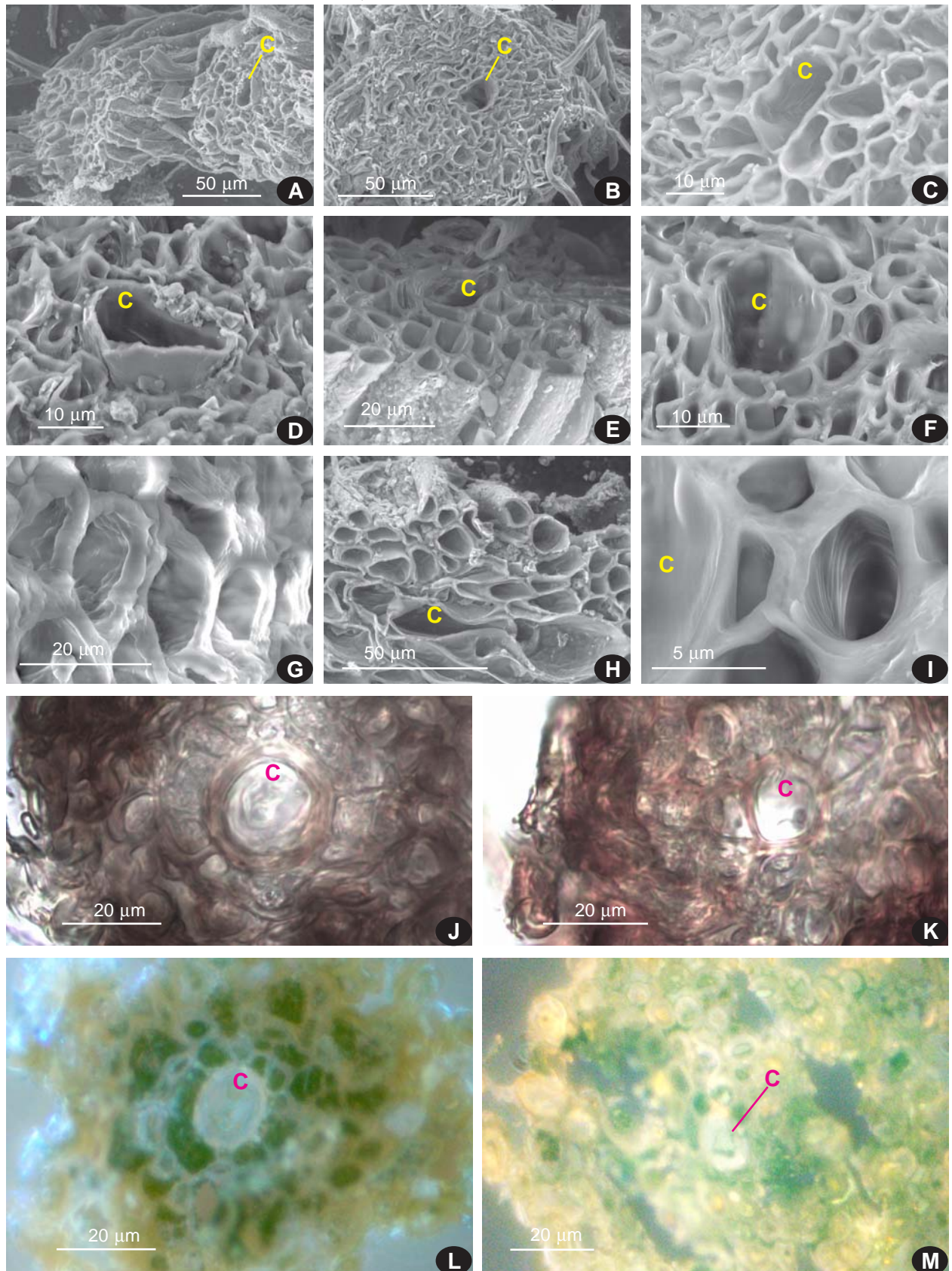


Fig. 9. Rhizoid rope structure of *Atrichum undulatum* (A, D, G, J, L) and *A. flavisetum* (B–C, E–F, H–I, K, M): razor blade sections under SEM (A–I), razor blade sections under light microscope (J–K) and microtome 1 μm sections under fluorescent microscope, autofluorescence excitation 330–385 nm. Note the caulonema (C) in the former species is thicker and having thicker cell wall and also that rhizoids are more strongly modified in the internal part of rhizoid rope than in the latter species (cf. L and M).

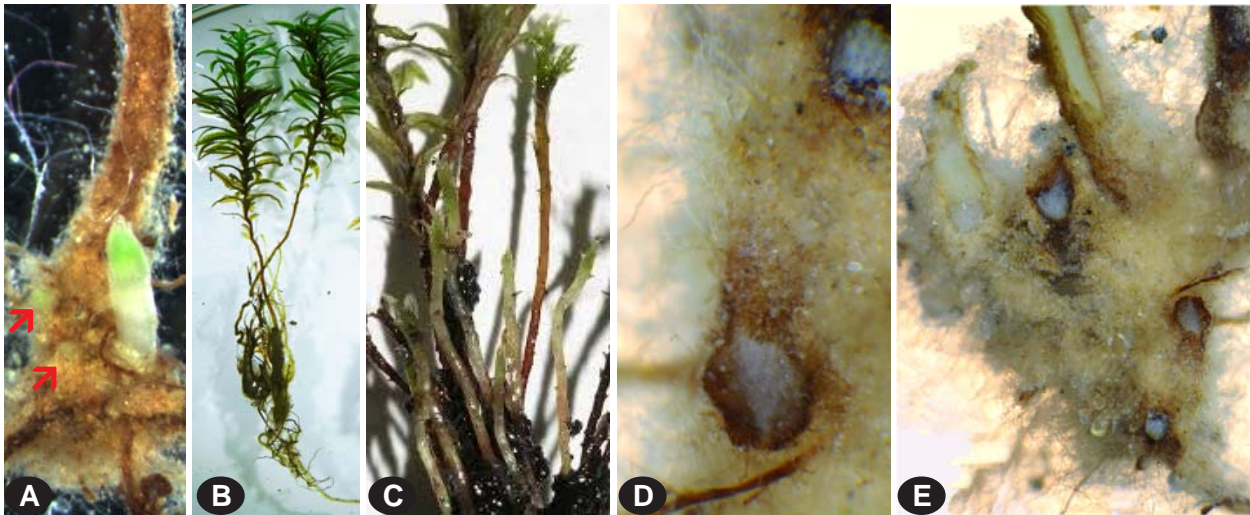


Fig. 10. Stem bases of *Atrichum undulatum* showing innovations at stem base (A, arrowed), rather dense stem bundles (B, C) and sublongitudinal section at base where several stems are fused, the lacunes shows partly decomposed stem bases within the callus-like tissue (D,E).

The comparison of the ordinary razor blade sections of *A. undulatum* and *A. flavisetum* is difficult, as the rhizoidal ropes in the latter are soft for obtaining cuttings good for observation. Fig. 9J and 9K show the wider caulonema in *A. undulatum*, which is seen in this species sometimes, but not always. The important, however, is that so wide caulonema has never been observed in any rhizoidal rope section of *A. flavisetum*.

The microtome 1 μm thick sections in Epon-Araldite medium studied for autofluorescence with ultraviolet excitation (Fig. 10C–D) show a stronger modification of rhizoids adjoining to the caulonemata. In *A. undulatum*, the wider caulonemata with more homogeneous cells around it were seen more often than in *A. flavisetum*, though there are exceptions, and, e.g., in Fig. 9I inner cells in *A. flavisetum* are also fairly regular.

The transverse sections show rather unusual ability of *Atrichum* parts to confluence in united structures. It seems that the stronger ‘confluence’ occurs in *Atrichum undulatum* than in *A. flavisetum*, and it therefore has a somewhat greater ‘rhizoidal rope force’, as was found in our tests with carrying weights.

Interestingly, some ‘confluence’ is occasionally seen in the stem base of *A. undulatum*, where innovations appear too close to the maternal stem, and may fuse, as shown in Fig. 10, on the plant collected at a trail side on a quite dense soil. During our studies such examples were not very numerous, but they appear occasionally, ensuring that *A. undulatum* more often forms bundles of several shoots (partly seen in the Fig. 2).

Certainly, observations of underground structures of these two species need further attention, as likely they yield in a better understanding of structural basis for the difference in their ecology.

Both *A. flavisetum* and *A. undulatum* belong to the the ‘short lived shuttle’ strategy (During, 1979), as the

age of first reproduction is about in 1–3 years. However, the sexual reproductive effort of *A. flavisetum* is obviously higher than in *A. undulatum* (Figs. 1–2), so the latter species by this trait is somewhat more similar to the ‘perennial shuttle’ strategy. This correlates well with habitat longevity of these species. Bare soil walls of upturns exist about 10–20(–25) years in this forest (sometimes less in case of very strong rains). Bare soil patches in flat places along the forest trails commonly occupied by *A. undulatum* are lasting much longer, several decades at least. The molehills are temporary, but starting growth on them, *A. undulatum* also survives long, judging from abundance of tufts in broad-leaved forests where the species persists in many flat places.

The division of habitats between two species seems a natural event, and the rarity of *A. flavisetum* in Moscow Province in previous century was likely a result of silvicultural usage that retained not enough old-growth forests in the region which are needed for this species.

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Supplementary Materials1: moving of rhizoidal rope data

https://kmkjournals.com/upload/PDF/Arctoa/33/Arctoa_33_226_238_table.xlsx

Supplementary Materials2: moving of rhizoidal rope video:

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