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ON THE DISTINCTION OF TWO SPECIES OF PALAEOZOIC MOSS GENUS *INTIA* (PROTOSPHAGNALES)

О РАЗЛИЧИЯХ ДВУХ ВИДОВ ПАЛЕОЗОЙСКИХ МХОВ РОДА *INTIA* (PROTOSPHAGNALES)

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Abstract

Upper Palaeozoic mosses of the genus *Intia* (order Protosphagnales) are widespread in deposits of Angaraland. The distinction of the two especially common species is in the focus of the study. The differences concern mostly leaf areolation pattern which is quite difficult to describe. The digitized areolation can be used for cell comparison by width to length ratio, allowing species discrimination in typical cases. Another way of their distinction is based on the ontogenetically-structured data. The latter is possible in Protosphagnalean mosses due to specific areolation pattern forming in the course of last cell divisions, so most cells are arranged in easily recognizable triads or tetrads. Proportions between cell areas within triads or tetrads appear to be a useful characteristic of species.

Резюме

Верхнепалеозойские мхи рода *Intia* (порядок Protosphagnales) широко распространены в отложениях Ангариды. В работе исследуются два особенно часто встречающихся вида, различия между которыми трудно поддаются словесному описанию, поскольку касаются, главным образом, рисунка клеточной сети листа. В ряде случаев для решения этой задачи оказывается достаточным соотнести ширину и длину клеток, что можно сделать с помощью разработанного метода оцифровки клеточной сети. Также хорошие различия выявляются при использовании онтогенетически структурированных данных. В случае протосфагновых мхов это возможно благодаря особому порядку деления клеток, в ходе которого образуются легко узнаваемые триады и тетрады. Соотношения площадей клеток внутри триад и тетрад оказываются существенной характеристикой вида.

KEYWORDS: cell divisions, fossils, *Intia*, leaf morphogenesis, mosses, Paleozoic, Permian, Protosphagnales

INTRODUCTION

The genus *Intia* was described by Neuburg (1956, 1960) for numerous moss remains in the Upper Paleozoic deposits of Angaraland. She described four species in this genus, all of them having characteristic oblique rows of laminal cells.

Two species, *Intia vermicularis* Neub., the type of the genus, and *I. variabilis* Neub., were found to be especially common. Two other species, *I. angustifolia* Neub. and *I. falcifolia* Neub., were represented by relatively few specimens differing mostly in leaf shape (Neuburg, 1960). The differences between the two former species include not only leaf shape, but also an areolation pattern, which was well illustrated, but described only in general: *I. vermicularis* has worm-like cells, while *I. variabilis* cells are more rounded.

Areolation patterns are often difficult to describe. Like verbal descriptions of ornaments, this is a very specific task. These things are much easier to illustrate than to express in appropriate words, and terminology of areolation patterns, including those in mosses, remains poorly developed. However, for identification of fossil mosses, this can be essential, as well-preserved fragments are usually quite small. At the same time variation within one lamina in Protosphagnalean mosses can be much greater than in modern ones, as different leaf parts of the former are developed in a somewhat independent way (Maslova *et al.,* 2012).

This differentiation problem is related to another one: the areolation pattern of *Intia vermicularis* may intergrade to that of the genus *Protosphagnum*, where narrow cells are much darker and cell dimorphism somewhat reminiscent of modern *Sphagnum. Protosphagnum*-like areolation is often seen in the proximal part of leaves, which in their distal part looks like perfect *Intia.* This fact forced Fefilova (1978) to describe the genus *Syrja-*

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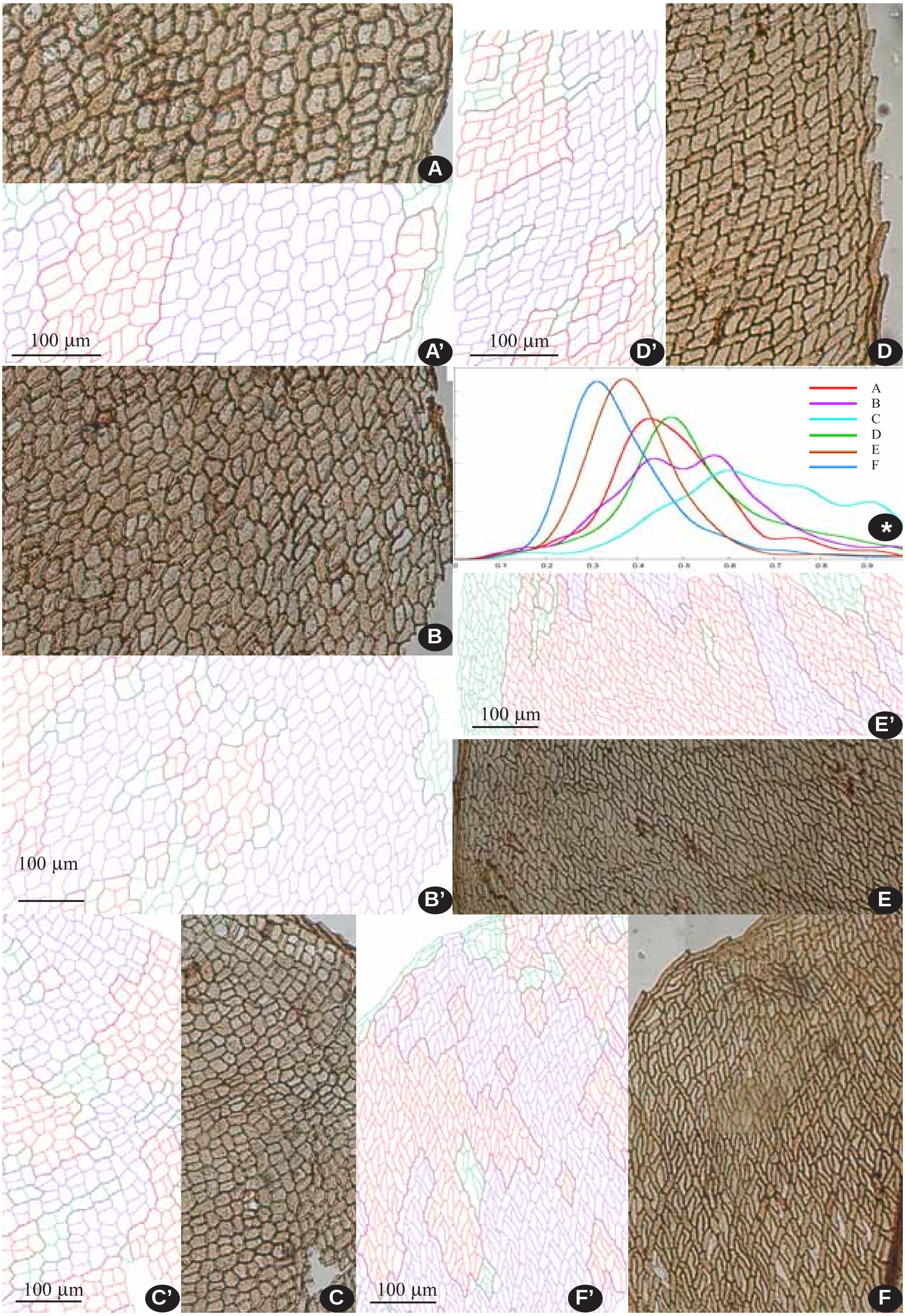
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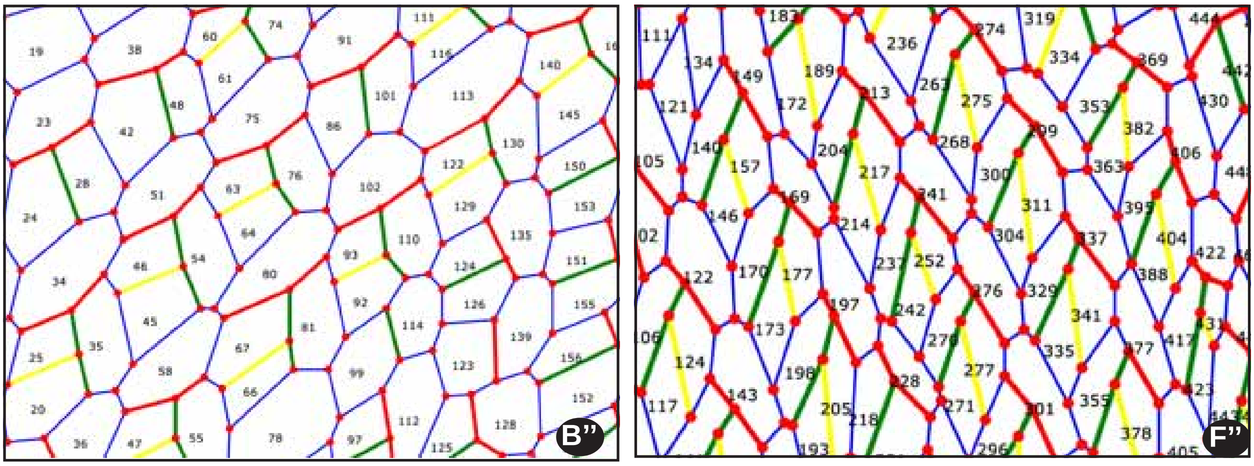
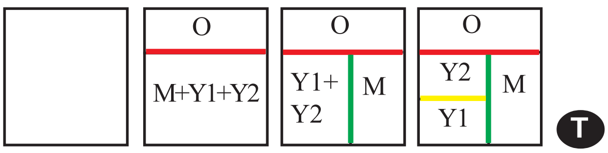


Fig. 2. Outinings of areaolation patterns B’ and F’ (cf. Fig. 1), with manual assignings of sequence of divisions according to scheme T, interpreting cells of triads and tetrads in term or younger sisters (Y), middle sister (M) and older sister (O).



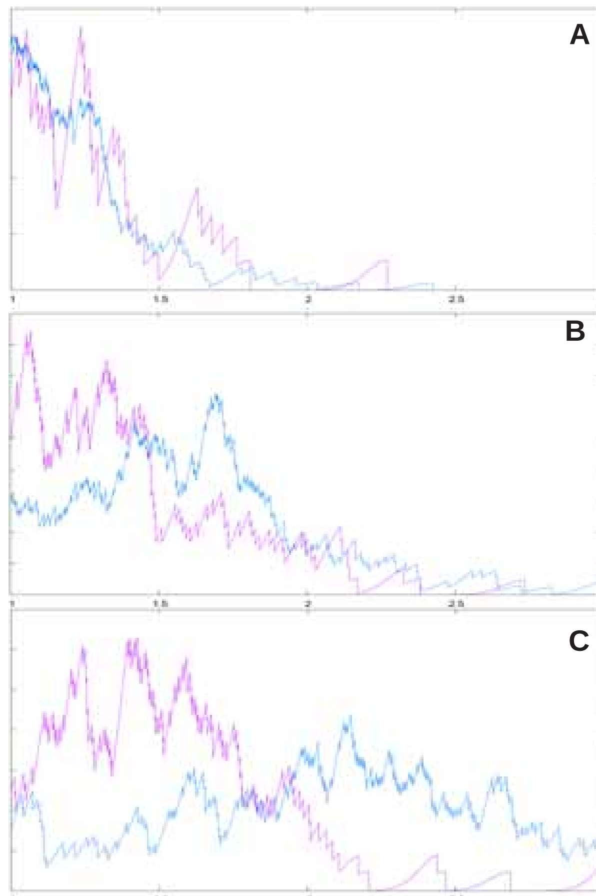


Fig. 3. Three distributions of proportions of cell areas in areolations of *Intia variabilis* (B’) in purple and *I. vermicularis* (F’) in blue (cf. Fig. 1*), with consideration of cell origin (cf. Fig. 2). Axis X in A: Y2:Y1; in B Y1+Y2:M; in C: M+Y1+Y2:O. Axis Y: arbitrary units. Note that separation of species by ontogenetically structured parameters gives better results than simple parameters of cells (cf. Fig. 1).

gia, characterized specifically by this combination of two areolation patterns. However, Ignatov (1990) suggested that this is a general rule, consequently the latter genus is superfluous.

At the same time, the areolation of *Intia variablis* intergrades not to *Protosphagnum*, but rather to *Vorcutannularia*, another genus of Protosphagnalean mosses (Maslova *et al.*, 2012), and likely *Intia vermicularis +Protosphagnum*, and *I. variabilis +Vorcutannularia* represent two different evolutional lineages.

Therefore, we undertook search for additional differentiation criteria of these two species of *Intia*, using the opportunities of statistical analysis of digitized areolation, previously applied to extant *Plagiomnium* species (Ivanov & Ignatov, 2011, 2012). This approach demonstrated interesting results in statistical evaluation of cell area, their linear parameters like length and width, arrangement of cells in oblique rows by means of analysis of massifs of thousands of cells. The usefulness of such approach was expected as *Plagiomnium* species are similar in many ways to Protosphagnalean mosses, including *Intia* (cf. Maslova et al., 2012).

METHOD AND MATERIALS

Additional collections discussed in this paper were gathered in Pechora Coal Basin (see details in Maslova et al., 2012). The laboratory preparation of material and specimen photography are given in the same publication as well.

Fig. 1 (opposite page). Lamina areolation of leaves of *Intia variabilis* (A-C) and *I. vermicularis* (D-F), with manual outlining of cells (corresponding letters with ’). Cells triads and tetrads arranged in oblique rows are in red, longitudinal in purple, uncertain in green. Scale bars 100 µm for C-F, 50 µm for A-B. Graph (*) shows the distribution of rate of cell width to length, given at axis X; axis Y – arbitrary units. Note that small area outlining from upper part of leaf (A and D) may not be enough for species differentiation (green and red lines).

Digitizing of laminal areolation for fossil plants is different from that used for modern ones (cf. Ivanov & Ignatov, 2011, 2012), as unfortunately the polarized microscopy can not be applied for fossilized specimens. Thus we simply outlined cells in graphic editors, as shown in Fig. 1. We used the photographs made at 10x objective magnification, several of them were combined together to obtain enough cells, so that the files were 5-12×3-8 thousand pixels, and cell outlining was done with a brush of 5 pixels wide. This allowed using a slightly modified version of outlining program described by Ivanov & Ignatov (2012).

While outlining cells for Fig. 1, we used two different colors for cell triads or tetrads arranged in oblique-transverse (in red) and longitudinal directions (in purple); triads/tetrads cells of uncertain direction were outlined in green. Direction is easily seen by conspicuous cell rows, or more precisely, by rows of "older sister cells", as defined in Fig. 2T. Such coloring allows a better view of triads and tetrads, and it also helps to understand the overall leaf development which will be discussed in a separate paper.

ANALYSES APPLIED

The visual comparison of areolation in Figs. 1A-C and 1D-F is consistent with the Neuburg (1960) descriptions of cells of *I. variabilis* as more rounded and of *I. vermicularis* as more elongate-vermicular.

The graph in Fig. 1* shows the distribution of cell width to length ratio, where these parameters are defined correspondingly as width and length of a minimal rectagular circumscribing a cell (Ivanov & Ignatov, 2011). As it is seen in the graph, the cells in *I. vermicularis* are generally longer than in *I. variabilis*, although with a considerable overlap (cf. graphs for Fig. 1B-C and Fig. 1E-F), and with poor differentiation between small areas taken from the upper part of a leaf of *I. vermicularis* (Fig. 1A and Fig. 1D).

ANALYSIS OF CELL AREA RATIOS

The motivation for additional study was connected with the characteristic T-pattern in areolation (Fig. 2T). We presumed that the distribution of cell area ratios may be useful. However, application of various autocorrelation functions was unsuccessful.

Finally we decided to use ontogenetically-structured data. This is possible in Protosphagnalean mosses due to the specific order of cell divisions where, in the course of the last divisions, most cells appear in easily recognizable triads or tetrads (Fig. 2T). If we take examples of areolation in Figs. 1B & B' and 1F & 1F', the last cell divisions in the corresponding leaves can be understood as shown in Figs. 2B'' and 2F''. The cell sorting into younger, middle and older sisters was manual, following the scheme in Fig. 2T.

Ratio of cell areas was used in the analysis. Fig. 3 shows that the areas of younger sisters in the two species are about the same (3A), while the ratio of combined areas of younger sisters to the area of middle sister is markedly larger in *I. vermicularis* than in *I. variabilis* (3B), and the ratio of areas of younger +middle sisters to older sister shows even more sound difference between these species (3C).

This result indicates fairly different ways of forming areolation at latest stages of leaf development. In *Intia variabilis,* the first division within the mother cell of triad/tetrad comes rather equally, and, after splitting off the older sister, the rest of the mother cell seems not to change in size, *i.e*. to develop like in scheme of Fig. 2T.

In *Intia variabilis,* however, the rest of mother cells continues enlarging after splitting off an older sister, thus middle and younger sisters become sufficiently larger in volume than the older one. It is unclear what is more important in this case: either unequal division or cell's ability to continue elongation, however, in favor of the latter is the fact that in a subapical part of leaf, the cell divisions are not especially unequal (Fig. 1D).

This example shows that even a partial involvement of ontogenetic data may sufficiently contribute to the comparative morphology.

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