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Taxonomic revision of the genus Cheilymenia – 6 Cheilymenia polaripustulata sp. nov. – a new species of the section Striatisporae

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Moravec J. (1997): Taxonomic revision of the genus Cheilymenia - 6. Cheilymenia polaripustulata sp. nov. - a new species of the section Striatisporae. - Czech Mycol. 50: 189-200

Cheilymenia polaripustulata sp. nov., belonging to the section Striatisporae J. Mor., ser. Tenuistriatae J. Moravec (1990, is described from Moravia, Czech Republic. The new species is distinguished by its smaller ascospores and by conspicuous cyanophilic pustules which are situated on the ascospore poles. The polar pustules, proved also by SEM photomicrographs demonstrated here, represent a unique feature in the genus. Relations of species of the series Tenuistriatae, their taxonomic value and position in the section Striatisporae and the delimitation of the section in the genus Cheilymenia is discussed. The author stresses that only the use of such a staining method which strictly avoids heating the slides, allows to recognize and examine the ornamentation of the separable perisporium of ascospores in Cheilymenia under the optical microscope. In an additional note, the author elucidates that the term "protype" (passed as Art 7.9bis), used in J. Moravec (1993) in the typification of Cheilymenia stercorea, should be substituted by the legitimate term epitype (Art.9.7, ICBN,1994).

Key words: Cheilymenia polaripustulata sp. nov., section Striatisporae, series Tenuistriatae, Discomycetes, taxonomy.

Moravec J. (1997): Taxonomická revize rodu Cheilymenia – 6. Cheilymenia polaripustulata sp. nov. – nový druh sekce Striatisporae. – Czech Mycol. 50: 189–200

Cheilymenia polaripustulata sp. nov., je popsána z Moravy v České republice, a zařazena do sekce Striatisporae J. Mor. ser. Tenuistriatae J. Moravec (1990). Nový druh je odlišen menšími askosporami a nápadnými cyanofilními bradavkami, umístěnými na pólech. Tyto polární bradavky, které jsou ojedinělým znakem v rodě Cheilymenia, byly prokázány též použitím elektronové mikroskopie a jsou zde demonstrovány SEM mikrofotografiemi askospor. Jsou diskutovány vzájemné příbuzenské vztahy a taxonomická hodnota druhů ser. Tenuistriatae a umístění a vymezení obou sérií i sekce Striatisporae v rodě Cheilymenia. Autor zdůrazňuje, že jedině použití správné metody barvení mikroreparátů, která se striktně vyhýbá ohřevu podložních skel s preparátem, dovoluje rozpoznat a zkoumat ornamentiku oddělitelného perisporu askospor u rodu Cheilymenia optickým mikroskopem. V dodatečné poznámce, autor upřesňuje, že termín "protype" jím použitý (J. Moravec 1993) při typifikaci Cheilymenia stercorea podle původního doporučení (Art 7.9bis) má být nahrazen konečným správným termínem epitype (Art.9.7, ICBN,1994).

The section *Striatisporae* J. Mor. was proposed in J. Moravec (1990) to accommodate species of the genus *Cheilymenia* which are characterized by yellow to orange-yellow apothecia with hairs possessing a bulbous, multituberous to obtusely rooting base in combination with a cyanophilic longitudinal rib-like

striation which covers the delicate separable outermost sheath (perisporium) of the ascospores. The section is divided into two series – ser. *Striatisporae* J. Mor. and ser. *Tenuistriatae* J. Mor.

Ser. Striatisporae is represented by two species, Cheilymenia theleboloides (Alb. et Schw.: Fr.) Boud. (the type species of the section) and Cheilymenia lemuriensis Le Gal., while ser. Tenuistriatae was created for Cheilymenia oligotricha (Karst.) J. Mor. [= Cheilymenia vitellina (Pers.: Fr.) Dennis, a correct name for the type species of the series], Cheilymenia pediseta (Clem.) J. Mor. and Cheilymenia citrinella (Velen.) Svr.

The delimitation of the two series in the section is particularly founded on three important characters: 1. – thickness of the ribs which form the perisporial ascospore striation; 2. – length and form of the apothecial hairs; 3. – differences in the medullary layer of the excipulum.

- ad 1. The separable ascospore perisporium of species of the ser. Striatisporae possesses a cyanophilic longitudinal striation similar to that of species of the section Coprobiae J. Mor., which is well visible under a lens of the optical microscope and consists of low longitudinal ribs which are often anastomosing and occasionally thickened or interrupted in the form of longitudinal chains of elongated warts [see Figs 19 a, b, c, and SEM Figs 31-32 in J. Moravec (1990)], whilst that of the ser. Tenuistriatae bears longitudinal ribs which are much finer and more densely arranged and hence hardly visible, observable only under an oil immersion lens, but proved by SEM and clearly scanned on SEM microphotographs [see Figs 20 a, b, c, and SEM Fig. 33 and Fig. 36 in J. Moravec (1990)].
- 2. The apothecial hairs of the species of the section, although seemingly of a different shape in each series, are in reality of the same type. We can find hairs with the same shape of their base as well as with various intermediate forms in both series. Nevertheless, the hairs of species in ser. Striatisporae are much scarcer or very sparsely distributed or even sometimes missing on the external surface and margin of the apothecia. They are short, hypha-like and flexuous to almost stiff and straight, superficial, thin-walled with walls up to 2 μ m thick, pale to yellow, often with a bulbous base [(see Figs. 7 a, b, in J. Moravec (1990)], whilst these of the ser. Tenuistriatae are in general much longer and stiffer, often pseudorooting with a bulbous, simple or multituberous base (hairs on the lower surface of apothecia) or with the obtusely rooting multifurcate base of rigid, thick-walled yellow-brown marginal and submarginal hairs [see Figs 8 a, b, c, in J. Moravec (1990)].
- 3. Concerning the construction of the apothecia, the ectal excipulum of both series (just as in all species of *Cheilymenia*) is composed of a textura globulosa to textura angularis. However, a delimiting feature is the structure of the medullary excipulum (medulla) which is only inconspicuously differentiated in the ser. *Striatisporae*, with only occasional, mostly inflated hyphae admixed into prevailing

angular cells (textura angularis to textura subintricata) in the medullary layer, whilst the medulla in the ser. *Tenuistriatae* consists of a textura subintricata to textura intricata, as such hyphae are denser and commonly present there [compare Fig. 2 to Fig. 3 in J. Moravec (1990) and see Fig. 1 of the present paper].

Regarding the individual species of the series *Tenuistriatae*, I consider worth adding some important notes including their taxonomy and nomenclature.

Due to the unclear typification of C. vitellina, particularly because Cooke (1876) illustrated Peziza vitellina Pers. with roughly ornamented ascospores, I considered P. vitellina a nomen dubium (J. Moravec 1990). This opinion was supported by the fact that the type material of Peziza vitellina * sabulosa Pers. from Mougeot's Herbarium, erroneously designated by Cooke as the type of Peziza vitellina (now deposited in K), possessed warted ascospores and refers to Scutellinia umbrorum (Fr.) Lamb. as proved by Schumacher (1988) and confirmed in J. Moravec (1990). Consequently, in the cited paper I proposed the name Cheilymenia oligotricha (Karst.) J. Mor., based on examination of the type of Peziza oligotricha Karst., to be used for the fungus commonly known as Cheilymenia vitellina. Now, I agree with Nannfeldt (1946) and Yao and Spooner (1996) that the type specimen of Peziza vitellina is that which comes from Persoon's herbarium under No 910, 261-823, and that Persoon's diagnosis of Peziza vitellina Pers. can be demonstrative enough for this fungus, and that the name Cheilymenia vitellina (Pers.:Fr.) Dennis can be a well established name for the discomycete in the currently accepted concept of the species with yellow apothecia and a terrestrial habitat.

C. vitellina is a very common species and was collected frequently by me in the Czech Republic and Slovakia. I have also examined a number of collections from other herbaria (K, PC, H, UPS, PAN, VELTU and others) and according to my examination, the ascospore size of C. vitellina is $(14-)15.5-18(-20) \times 7.5-$ 10.5 μ m (mostly 17 × 8.5 μ m). For instance, ascospores of the type (K) of Peziza dalmeniensis Cooke [= Cheilymenia dalmeniensis (Cooke) Boud. = syn. of C. vitellina measure 14.5-17(-18.5) \times 7.5-9.7(-10.2) μ m, those of the type of Peziza oligotricha P. Karst. [= Cheilymenia oligotricha (P. Karst.) J. Mor. = syn. of C. vitellina] measure 15-18.5(-20) \times 8-10.5 μ m. The apothecial hairs of C. vitellina, are notably long $-200-750-1000 \times 10-24-30 \,\mu\mathrm{m}$ with walls 0.5-3 $\,\mu\mathrm{m}$ thick. The paraphyses are slightly enlarged (up to 6 μ m) at their apex. It is a species of a terrestrial habitat found mostly on forest paths, and its frequent occurrence under Urtica dioica mentioned by Svrček (1947) is really conspicuous. The apothecia fructify on soil and also on debris of putrid twigs and among moss under Urtica without any sign of pollution by excrements and according to my opinion and experiences, C. vitellina is a strictly "terrestrial", non-coprophilous discomycete which may be confined to Urtica or incidentally to other plants by a kind of symbiosis.

Two other species, Cheilymenia pediseta (Clem.) J. Moravec (1990) and Cheilymenia citrinella (Velen.) Svrček (1977) were considered to be members of this series and distinct from C. vitellina. Nevertheless, after further examination of many collections of C vitellina, in respect to its certain, though modest variability, I have concluded that these two species differ only slightly from C. vitellina, which may be obvious from the following comments:

A part of the type collection of Sepultaria pediseta Clem. (a section of an apothecium mounted on a slide covered by cover glass with "Lachnea pediseta" written on it), Nebraska, on horse dung, Sept. 4, 1895 leg. F. E. Clements, marked "type" (NY) is packed together with a specimen of Cheilymenia coprinaria (Cooke) Boud. and this may explain why Denison (1954) synonymized Lachnea pediseta (Clem.) Sacc. with C. coprinaria. The hairs of the type (of the section on the slide) measure $100\text{-}675 \times 10\text{-}30~\mu\text{m}$ and are thus shorter than but of the same type as those of C. vitellina. [compare Figs. 8 b and 8 c in J. Moravec (1990)]. The ascospores measure $15.5\text{-}19(\text{-}22) \times 8\text{-}9~\mu\text{m}$ and so they are somewhat more elongated than those of C. vitellina. In addition, the paraphyses are not or only very slightly enlarged (up to $4.5~\mu\text{m}$). These differences, including the true coprophilous habitat of C. pediseta, has prevented me from a satisfied conclusion that it is merely a form of C. vitellina.

Analogously, also C. citrinella is very close to C. vitellina. The holotype of Lachnea stercorea var. citrinella Velenovský (1934), Bohemia, Struhařov prope Mnichovice, on cow dung, VIII. 1925 leg. Velenovský (PRNM 147289) consists of a single apothecium only and the hairs are of the same type as those of C. vitellina, including shape of their bases [see Fig. 8 a in J. Moravec (1990)] and the thickness of their walls (1-3 μ m), but they are conspicuously shorter as they measure $120\text{-}280 \times 12\text{-}29 \ \mu\text{m}$ [250-350 \times 12-17 μ m according to Svrček (1949)] and are absent from the margin of the lemon-coloured apothecium. The ascospores measure $(15.5\text{-})16.5\text{-}19.5(\text{-}21.5) \times (7.5\text{-})8\text{-}9(\text{-}10.5) \ \mu\text{m}$ and the paraphyses are apically not or only very slightly enlarged $-3\text{-}3.5(\text{-}4) \ \mu\text{m}$. These differences as well as the growth on dung may sufficiently distinguish C. citrinella from C. vitellina. It is, however, closer to C. pediseta particularly due to the coprophilous habitat, elongated ascospores and inconspicuously enlarged paraphyses, but it differs by much shorter hairs which are absent from the margin of the apothecia.

Another species which increases the number of species of the section Striatisporae ser. Tenuisriatae is described as a new taxon here.

Cheilymenia polaripustulata J. Moravec, sp. nov.

Figs 1-4.

Apothecia 4-7 mm diam., sessilia, profunde patellaria, margineque undulata vix lobata et pilis brevibus vel medio-longis luteo-fuscis, sparsis obsita, extus et

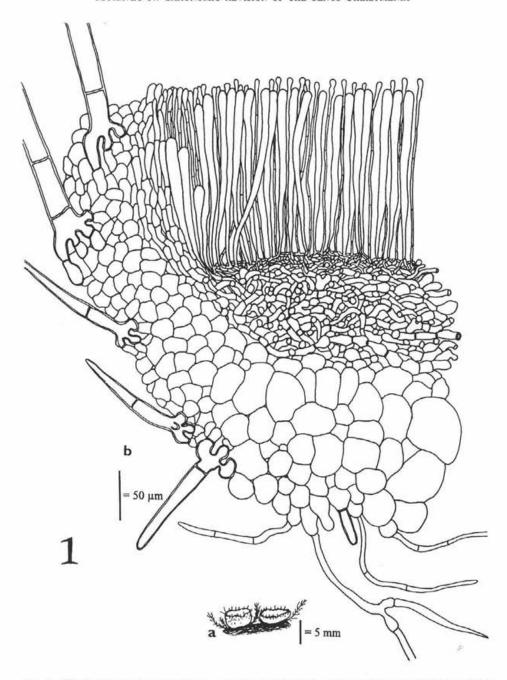
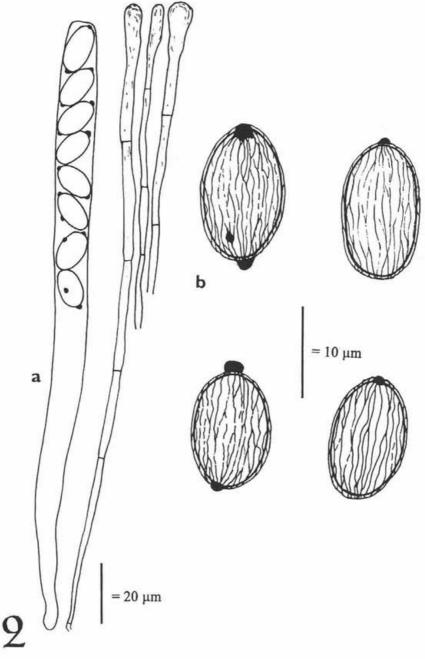
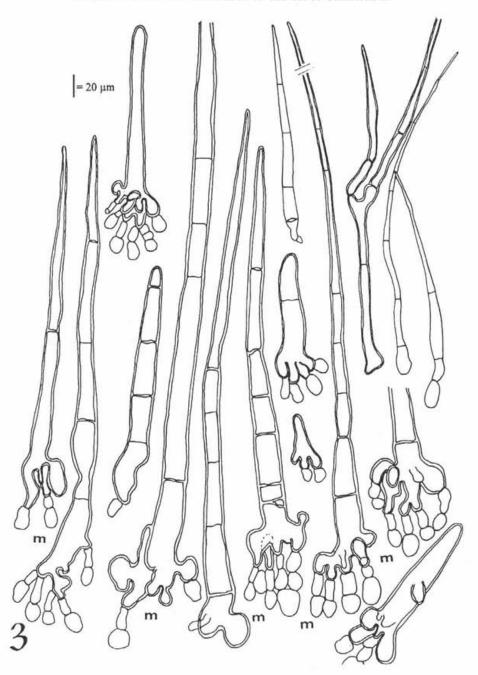


Fig. 1. Cheilymenia polaripustulata sp. nov.: a. apothecia; b. section of the marginal part of the apothecium. Holotype BRNM.



 $\begin{tabular}{ll} {\bf Fig.~2.~Cheilymenia~polaripustulata~sp.~nov.:~a.~ascus~and~paraphyses;~b.~ascospores~(oil~immersion).~Holotype~BRNM. \end{tabular}$



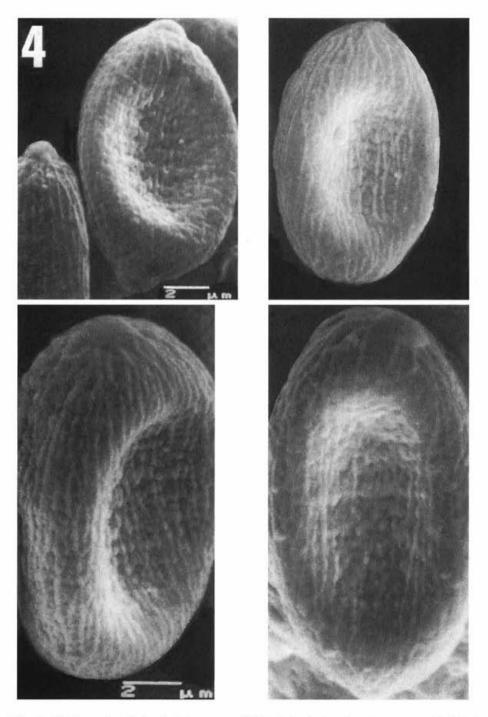
 $\label{eq:Fig. 3. Cheilymenia polaripustulata} \ \text{sp. nov.: apothecial hairs (m = marginal hairs)}. \ \text{Holotype BRNM}.$

in hymenio luteo-aurantiaca, parte externa pilis brevibus luteolis crebre obsita. Excipulum externum e textura globuloso-angulari. Excipulum internum (medulla) e textura subintricata usque intricata. Pili 60-540 \times 12-27 μm , basi tuberosi, multituberosi, subradicantes, usque bulboso-radicantes et bulboso-furcati vel obtuse multifurcati, apice obtusi vel subacuti, septati, crasse tunicati, luteo-brunnei. Asci 180-210 \times 9.5-10.5 (-12) μm , cylindracei, octospori. Ascosporae ellipsoideae, (11-) 12-15 (-16.5) \times 7.5-9 (-9.7) μm (plerumque 13.6 \times 8 μm), perisporio separabile, subtiliter dense longitudinaliter striato cum costis cyanophilis, tenuibus et humilibus, irregulariter anastomosantibus et saepe cum pustulis magnis, cyanophilis ad polis donatae. Paraphyses filiformes, 3.5-4 μm crassae, apice sensim vel clavato-incrassatae (4-9 μm).

Habitat: Moravia, Třebíč, ad terram humosam cum excrementa gallinaria mixta, inter muscos ad viam in horto in urbe, 10. VIII. 1986 leg. Jiří Moravec. Holotypus BRNM, duplicatum in herbario privato J. Moravecii (J. Mor.) asservantur.

Apothecia 4-7 mm diam., sessile, cupulate, yellow-orange with an undulate or nearly lobed margin; the margin and outer surface are sparsely covered with inconspicuous, short to average-sized hairs. Excipulum differentiated. Ectal excipulum of a textura globulosa-angularis, composed of globose, subglobose to angular cells which are 15-45-65 μm in diam. Medulla of a textura subintricata to textura intricata composed of angular to elongated cells which are intermixed with hyphoid cells or septate, often inflated interwoven hyphae 4-7.5 μ m in diam. Hypothecium of a textura subintricata of smaller cells and hyphal elements. Hairs $60-540 \times 12-27 \mu m$, those on the lower surface of the apothecia are superficial, with a tuberous base, usually thin-walled and pale yellow, the marginal hairs yellow to yellow-brown, straight, rigid, septate, with obtuse or almost pointed apex, thick-walled (the walls 1-4 μ m thick), with tuberous to multituberous or widely, obtusely rooting, multifurcate base. Asci $180-210 \times 9.5-10.5$ (-12) μ m, cylindrical, with a blunt apex, eight-spored, operculate. Ascospores (11-) 12-15 (-16.5) \times 7.5-9 (-9.7) μm (mostly 13.6 × 8 μm, only spores developed in 4 – 6-spored asci may reach up to $16.5 \times 9.7 \,\mu\text{m}$), ellipsoid, eguttulate, hyaline, with a yellow refractive colour when stained with CB, with a separable delicate outer sheath which is covered by a longitudinal, very fine cyanophilic rib-like striation consisting of thin and very low ribs which are densely arranged and occasionally anastomosing, the fine ribs converge on the ascospore poles of the sheath and concentrate into large and conspicuous cyanophilic pustules situated on one or on both poles. Paraphyses filiform, 3.5-4 μ m thick, with a slightly enlarged or more clavate apex (4-9 μ m), septate, with a yellowish content.

Habitat: Třebíč, Czech Republic (Moravia), on moist mineral-rich soil mixed with hen excrements among dense moss cover on a path between a garden and



 $\textbf{Fig. 4.} \ \textit{Cheilymenia polaripustulata} \ \text{sp. nov.: SEM photomicrographs of ascospores. Holotype BRNM.}$

houses in the suburb of the town, 10. VIII. 1986 leg. Jiří Moravec. Holotype BRNM, isotype in herb. J. Mor.

C. polaripustulata is a natural member of the section Striatisporae, ser. Tenuistriatae discussed above. It is undoubtedly close to C. vitellina, especially due to the similar colour of the hymenium, the same type of apothecial construction, the same type of apothecial hairs, the shape of their base, and for the fine rib-like striation of the separable outer ascospore sheath (perisporium). It differs, however, by a smaller ascospore size, and by the presence of the strongly cyanophilic large pustules on the perisporium, which are situated on one or both ascospore poles and where the fine longitudinal ribs concentrate. The latter feature is unique in the genus and has never been seen by me on the ascospore perisporium of other species of Cheilymenia. It has been proved also by SEM prepared on samples taken directly from the dried hymenium without any treatment of ascospores by any liquid (SEM Fig. 4.). As proved after examinations of many collections, the fine ribs in C. vitellina converge on the ascospore poles but do not form any abscesses. Moreover, the ribs on the perisporium of C. polaripustulata are slightly thicker and more irregular, often irregularly interrupted and more anastomosed, and thus the striation is intermediate between that on the ascospore perisporium of species of the ser. Striatisporae and that of the ser. Tenuistriatae. Nevertheless, the striation in the ser. Striatisporae is formed by even thicker and not so densely arranged ribs which can be occasionally thickened or interrupted forming longitudinal chains of elongated warts, but never forming such large pustules on the ascospore poles. [Compare all cited SEM Figs. in J. Moravec (1990)]. Also, the paraphyses of C. polaripustulata are often clavate above, thus being more enlarged than those of other species of the series.

Cheilymenia lemuriensis Le Gal (1953) known merely from the type collection from Madagascar (also examined) possesses even smaller ascospores bearing much coarser ribs, and also for all its other features, belongs in the series *Striatisporae*. [see illustration in Le Gal (1953) and J. Moravec (1990)].

On this occasion, I wish to stress here the importance of using the proper staining method in which the slides are not heated, since this is necessary for examination under the optical microscope of the original ascospore ornamentation which covers the very delicate outermost sheath (perisporium) of the ascospores in *Cheilymenia*.

The original ornamentation of the separable outermost sheath of ascospores in *Cheilymenia* has usually not been recognized by even recent authors. This is obviously caused by using a wrong staining method, especially when lactophenol is used and the slides with micro-sections are heated. Such drastic treatment, as well as any violent way of dehydration of the apothecia usually destroys the very delicate separable outermost sheath of the ascospores (perisporium) – the

sheath can easily be damaged, deformed and consequently secondary wrinkled, or completely separated from the proper ascospore wall, and thus the ascospores may appear smooth or with secondary artefacts. As stressed in J. Moravec (1989a, 1989b,1993), the original cyanophilic ornamentation on the perisporium of ascospores in *Cheilymenia* is clearly seen and well recognizable under the optical microscope when cotton blue, which stains promptly without heating the slides, is used (CB Geigy s 123). The perisporial ascospore ornamentation in *Cheilymenia* has also been proved by SEM (J. Moravec 1987), but also the SEM samples must be taken only from the dried hymenium or ascospores spontaneously released from it may directly be coated by gold, and these must not be treated with any aggressive liquid, for the same reason as mentioned above.

Yao and Spooner (1996) mentioned that the British material of *C. vitellina* showed "a fine punctate ornamentation on some ascospores whilst the others are smooth". This observation must have been caused by using the wrong staining method. In fact, there are no punctations on the outermost sheath of the ascospores of *C. vitellina*, but some secondary artefacts can be seen (also on the epispore or endospore!) after the ascospore perisporium is damaged or separated by heating the slides. The fine rib-like striate ornamentation on the non-deformed and tense ascospore perisporium of *C. vitellina* is well seen by using an oil immersion lens when the sections are correctly stained without heating the slides, and of course much more clearly scanned on SEM photomicrographs.

Besides, Yao and Spooner (1996) have mentioned that ascospores of *C. vitellina* were illustrated in J. Moravec (1990) as having a "punctate-striate" ornamentation. This statement was obviously caused by a wrong interpretation of the figures in my paper. As is evidently seen in the line drawings in Fig. 20 a, and SEM Figs 33 and 36 in J. Moravec (1990), the illustration of the ascospores of *C. oligotricha* (= *C. vitellina*) clearly show the very fine longitudinal striation as discussed above.

Even if commonly neglected and underestimated, the perisporial ascospore ornamentation is one of the very significant characters important in the delimitation of species and infrageneric taxa of the genus *Cheilymenia* and correlates with other features of species of individual sections and thus represents one of the leading characters used for the infrageneric classification in J. Moravec (1990).

ADDITIONAL NOTE

Regarding the type species of the genus *Cheilymenia* and simultaneously the type species of the typical section (sect. *Cheilymenia*), *Cheilymenia stercorea* (Pers.:Fr.) Boud., I wish to add an important note on the nomenclature and typification.

Having rejected Denison's (1964) lectotypification of *Peziza stercorea*, I chose (J. Moravec 1990), on kind advice of Prof. R. P. Korf (Ithaca), Bulliard's figure of *Peziza ciliata* Bull., Herb. France 109: t. 438, f. 2, 1790, cited by Fries in his sanctioning work (Fries, Systema Mycologicum 2: 87, 1822), as illustrative of *P. stercorea* Pers.:Fr., to be the LECTOTYPE of *P. stercorea* Pers.:Fr. Simultaneously, as that illustration is demonstrably ambiguous, I designated (J. Moravec 1993) a specimen from the S herbarium which comes from Sweden, the country of Fries to be the "PROTYPE" to support that lectotype illustration, as provided under Article 7.9bis at that time.

In the cited paper we used the term "protype" as originally proposed for adoption by the International Botanical Congress in Tokyo in 1993 and made it clear that "protype" was a provisional designation of the term that was passed as the "Art 7.9bis (which may bear a different designation when the code is published)", becoming Art.9.7, ICBN, 1994. The final term adopted for "protype" by the Editorial Board of the Code is "epitype", and thus readers should substitute the term epitype where ever we used "protype" in J. Moravec (1993), and the selected collection from the S herbarium should be designated as an epitype of Peziza stercorea Pers.: Fr.

In this respect, the selection of another "epitype" made by Yao and Spooner (1996) has been superfluous as the authors do not take into consideration the reasons we stressed earlier (J. Moravec 1993). Moreover, in my opinion the type of *Humaria alpina* Fuckel designated by the cited authors as "holoepitype" of *C. stercorea* cannot serve for such typification since it represents a distinct form – f. alpina (Fuck.) J. Moravec (1990) – which differs from the typical form f. stercorea, as stated in J. Moravec (1993).

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