P E R S O O N I A Published by the Rijksherbarium, Leiden Volume 13, Part 1, pp. 89-96 (1986)

## NOTES ON CUP-FUNGI-3 On three species of Cheilymenia

## J. VAN BRUMMELEN

Rijksherbarium, Leiden

Three coprophilous species of *Cheilymenia* resembling *Lasiobolus* are redescribed. *Cheilymenia raripila* is reported from Germany; *C. insignis* and *C. pulcherrima* are newly described from authentic material of the Crouan brothers. *Cheilymenia hyalochaeta* is considered to be a synonym of *C. raripila*.

Cheilymenia raripila (Phill.) Dennis.—Fig. 1

Ascobolus raripilus Phill. in Grevillea 7: 23. 1878. — Lasiobolus raripilus (Phill.) Sacc., Syll. Fung. 8: 537. 1889. — Patella raripila (Phill.) Seav., N. Am. Cup-fungi (Operc.) 173. 1928. — Cheilymenia raripila (Phill.) Dennis in Kew Bull. 14: 428. 1960.

Peziza fraudans P. Karst. \* P. hyalochaeta Speg. in Anales Soc. cient. argent. 10: 24. 1880. — Neotiella fraudans (P. Karst.) Sacc. \* P. hyalochaeta Speg., Sacc., Syll. Fung. 8: 191.1889. — Cheilymenia hyalochaeta (Speg.) Gamundi in Lilloa 30: 326. 1960.

Apothecia gregarious to closely crowded, superficial, sessile 1-1.5 mm diam., 0.4-0.8 mm high. Receptacle cup-shaped to saucer-shaped, rather pale yellowish to yellowish brown; surface covered with only a few rather inconspicuous pale brownish or almost hyaline hairs; margin scarcely differentiated, but sometimes visible as a very narrow irregular, membranous collarette. Disc slightly concave to flat, even, at first deep yellow, then paler. Hymenium up to 210 µm thick. Hypothecium scarcely differentiated, of thin-walled isodiametric cells  $6-10 \ \mu m$  wide. Flesh up to 70  $\mu m$  thick, of polygonal or oblong thinwalled cells,  $14-25 \times 10-20 \ \mu m$  (textura angularis to globulosa) and hyphae  $6-10 \ \mu m$ wide lying at a low angle with the surface of the receptacle. Excipulum  $20-70 \,\mu m$  thick, near the base 40–70  $\mu$ m thick, at the margin 20–35  $\mu$ m wide, yellowish, consisting of angular or rounded, isodiametric to oblong, slightly thicker walled cells  $20-45(-65) \times$  $20-35 \,\mu m$  (textura globulosa to angularis), covered with superficially implanted, isolated hairs. Hairs of a single type, superficial, non-rooting, inflated at the base, simple, 0-5septate, straight, thick-walled (up to 2.5  $\mu$ m), 100–280  $\mu$ m long by 11–20(–25) $\mu$ m wide at the base, with hyaline or pale brownish walls, with rounded or, very rarely, pointed apices. Asci subcylindrical with a short stalk, rounded above,  $180-205 \times 24-26 \mu m$ , 8-spored; the wall not staining blue with iodine. Ascospores uniseriate, ellipsoid (length/ width ratio 1.7-2.0, average 1.9), hyaline  $(21.5-)23.0-26.0(-27.0) \times (12.5-)13.0-$ 14.5(-15.0) µm, without oil globules, smooth, surrounded by an easily loosening and wrinkling secondary spore wall. Paraphyses septate, slender, cylindrical, clavate at the end, unbranched,  $2.5-3.7 \mu m$  thick, enlarged up to 9  $\mu m$  at the tip, containing yellowish orange granules of pigment, especially in the upper part.

Habitat.—On cow dung.

Specimen examined.—GERMAN FEDERAL REPUBLIC, Bavaria, near Coburg, 20.X.1983, B. Hanffs.n. (L).

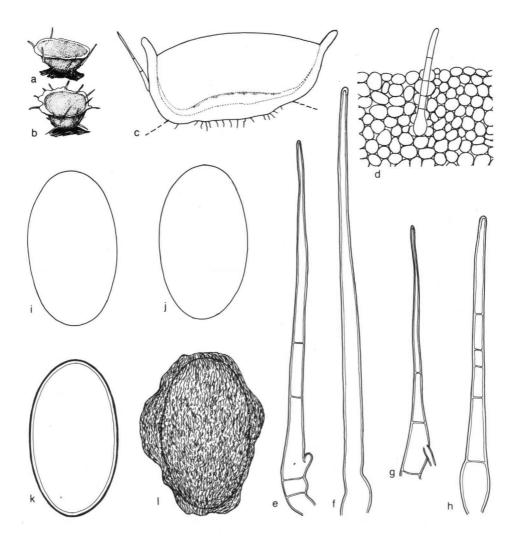


Fig. 1. Cheilymenia raripila (leg. B. Hanff). — a, b. Habit of fruit bodies  $\times 20$ . — c. Diagrammatic section of fruit body  $\times 63$ . — d. Texture of excipulum seen from outside  $\times 160$ . — e-h. Hairs  $\times 400$ . — i, j. Ascospores  $\times 1600$ . — k. Ascospore in optical section  $\times 1600$ . — 1. Ascospore with loose secondary wall (stained with methyl blue in lactic acid)  $\times 1600$ .

Apparently this is an uncommon species with a wide distribution. It is known from California (Phillips, 1878), Iowa (Seaver, 1905), Venezuela (Dennis, 1960), Argentina (Gamundi, 1975), Australia (Rifai, 1968), and the British Isles (Dennis, 1972, 1979, 1981; Hawksworth, 1976; Clarke, 1980; Kirk & Spooner, 1984). It has not previously been reported from the European Continent.

At first sight, this fungus may be taken for a species of *Lasiobolus* Sacc. because of the non-rooting inflated hair-bases that originate superficially from the outer cell-layer of the excipulum. These hairs, however, perfectly agree with the 'superficial hairs' distinguished by Denison (1964) in his study of the North American species of *Cheilymenia* Boud. According to this author such hairs predominate on apothecia of the 'theleboloides type'.

Several other important characters of this species favour the view of a position in the genus *Cheilymenia* as proposed by Dennis (1960).

The asci are not protruding above the surface of the hymenium at maturity as they do in *Lasiobolus*.

The fruit bodies are paragymnohymenial as in species of *Cheilymenia*. In such fruit bodies no closed sheath is formed over the ascogonium or the hymenium. In *Lasiobolus* the fruit bodies are cleistohymenial and open in the late mesohymenial phase, i.e. not before the ascospores are ripening (van Brummelen, 1967, 1972; Bezerra & Kimbrough, 1975).

The margin of the receptacle often ends in a short hyaline collarette, a phenomenon rather common in certain species of *Cheilymenia*, but unknown in *Lasiobolus*.

The terminal elements of the paraphyses contain many granules of a carotenoid yellowish orange pigment, not present in species of *Lasiobolus*.

The loosening and wrinkling of the otherwise smooth outermost layer of the ascospore wall as an envelope or sheath, when material is heated in lactic acid, is considered a diagnostic feature of the genus *Cheilymenia* (Le Gal, 1953; Denison, 1964). When stained with methyl blue in lactic acid or lacto-phenol the loose ascospore layer seems to show a fine net-work, because of the fine folds standing out from the surface.

*Cheilymenia raripila* is characterized by sparsely placed superficial, short, non-rooting, blunt hairs, relatively large ascospores, and a crowded growth of its fruit bodies on cow dung or rabbit pellets.

From Gamundi's (1960) detailed description of *Cheilymenia hymenochaeta* (Speg.) Gamundi after Spegazzini's original specimen it is evident that this name is a synonym of *C. raripila*.

Cheilymenia insignis (Crouan) Boud.—Fig. 2

Ascobolus insignis Crouan in Annls Sci. nat. (Bot.) IV 10: 196 pl. 13 H f. 38-43. 1858. — Humaria stercorea var. insignis (Crouan) Quél., Ench. Fung. 286. 1886. — Lachnea insignis (Crouan) Sacc., Syll. Fung. 5: 181. 1887. — Cheilymenia insignis (Crouan) Boud., Hist. Class. Disc. Eur. 63. 1907. — Dasyobolus insignis (Crouan) Le Gal in Annls Sci. nat. (Bot.) XII 1: 455. 1961. — Holotype: [Finistère, France] s. loc., on cow dung, XII.1857, Crouan (CONC-A2394).

Apothecia solitary or gregarious, superficial, sessile, 0.6-1(-4') mm diam., about 0.8 mm high (hairs included). Receptacle at first subglobular to slightly ovoid or urceolate, then hemispherical, yellowish orange; surface covered with many red-brown hairs, often apparently two rows of long hairs near the margin and a few short ones lower down; margin smooth or somewhat crenulate, often ending in a narrow, hyaline collarette. Disc flat or slightly concave, even, bright yellowish orange. Hymenium up to 300  $\mu$ m thick.

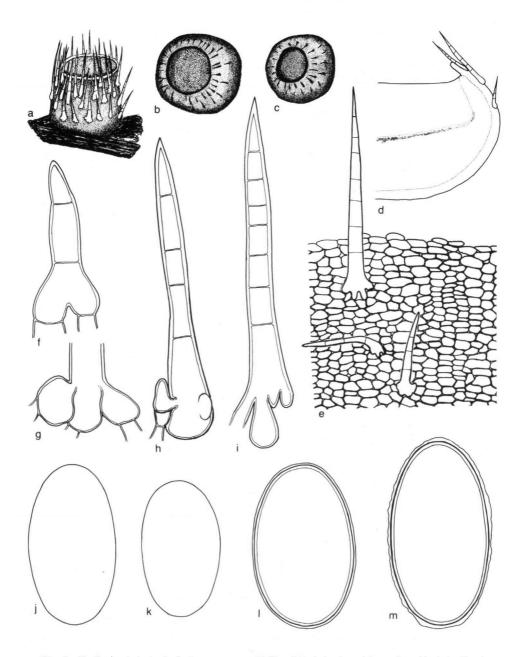


Fig. 2. Cheilymenia insignis, holotype. — a. Habit of fruit body  $\times 25$ . — b, c. Fruit bodies (redrawn after original drawings in Crouan herbarium, CONC). — d. Diagrammatic section of fruit body  $\times 40$ . — e. Texture of excipulum seen from outside  $\times 160$ . — f–i. Short hairs  $\times 400$ . — j, k. Ascospores  $\times 1600$ . — l, m. Ascospores in optical section (stained with methyl blue in lactic acid)  $\times 1600$ .

Hypothecium not clearly differentiated as a distinct layer. Flesh  $300-500 \,\mu m$  thick, of isodiametric to oblong thin-walled cells  $25-70 \times 25-55 \mu m$  (textura angularis to globulosa), yellowish. Excipulum clearly differentiated, 55-80 µm wide, hyaline, consisting of vertical rows of polyhedral thick-walled cells  $27-70 \times 23-40 \,\mu\text{m}$  (textura angularis). Hairs of a single type, arising from the two outermost cell layers of the excipulum, 2-18septate, straight, very brittle, thick-walled  $(1.8-3.7 \ \mu m)$ ,  $(100-)140-400 \ \mu m$  long by  $25-37 \mu m$  wide at the base, strongly forked and rooting at the base; roots partly inflated like vesicles; apices acuminate, rather sharply pointed, rarely blunt. Asci cylindrical with a short stalk, rounded above,  $250-290 \times 22-27 \mu m$ , 8-spored, not staining blue with iodine. Ascospores uniseriate, ellipsoid, rather variable in shape and size (length/width ratio 1.4-2.0, average 1.8), at first hyaline, with yellowish contents at maturity, (22.1-)  $23.0-32.0(-36.4) \times (12.6-)13.2-17.3(-18.5) \mu m$  (mean size  $27.5 \times 15.7 \mu m$ ), without oil-globules or granules, smooth; the outermost layer staining blue with methyl blue, only occasionally separating from the spore in lactic acid. Paraphyses septate, cylindrical, branched,  $3.5-5 \,\mu m$  thick, enlarged (up to 8  $\mu m$ ) or diverticulate at the tip, containing orange pigment granules, especially in the upper part.

Habitat.—On old cow dung.

Specimens examined.—FRANCE, Finistère: s. loc., XII.1857, Crouan (CONC-A2394, type of Ascobolus insignis Crouan); s. loc., 8.II.1862, Crouan (CONC-A2380); s. loc., 4.II.1869, Crouan (CONC-A2379).

The three specimens in the Crouan's herbarium represent the same species, although there is a considerable variation in the size of the ascospores. In the type specimen, the spores measure  $22.1-27.5 \times 12.6-17.6 \mu m$ , while they are larger in the other specimens. In addition to the herbarium specimens there are at least two original water-colour drawings by the Crouans of this species, which fully agree with the published drawings (Crouan, 1858: pl. 13H). These coloured drawings clearly show the bright yellowish orange contents of the paraphyses and the vividly orange colour of the disc.

No evidence could be found for the presence in this species of ascospores similar to those found in the genus *Ascobolus* Pers.: Fr., as described by Le Gal (1961: 454, fig. 5B).

As stated before (van Brummelen, 1967: 222), this species should be placed in *Cheilymenia* Boud. Especially the flat disc, the carotenoid pigment in the paraphyses, the strongly lobed excipular hairs, and the absence of the staining of the ascus wall with iodine are in favour of such a position.

Mature ascospores show rather homogeneous yellowish contents. With iodine a rather pale, but positive, red staining of these contents could still be observed. This indicates the presence of glycogen, a substance showing greenish opalescence with the earlier methods of microscopy using rather diffuse day-light. This may explain why the Crouans described the ascospores as glaucous green ('vert glauque').

Cheilymenia insignis is a rare species. Only Boudier (1869: 257), Fuckel (1870: 289), and Rehm (1895: 1055) give new records of it for respectively Montmorency (France), Oestrich (German Federal Republic), and Leipzig (German Democratic Republic), but apparently no material has been preserved by them.

It differs from *C. fimicola* (de Not. & Bagl.) Dennis in the larger ascospores and the more frequent excipular hairs.

From C. stercorea (Wigg.: Fr.) Boud. it can be distinguished by its larger ascospores and the absence of stellate hairs at the base of the fruit body.

It shows some similarity to C. pulcherrima, but differs in having fruit bodies of a different shape, hairs lobed at their base, and larger ascospores.

## Cheilymenia pulcherrima (Crouan) Boud.-Fig. 3

Ascobolus pulcherrimus Crouan in Annls Sci. nat. (Bot.) IV 10: 196. 1858. — Peziza pulcherrima (Crouan) Cooke, Mycographia 84. 1876. — Humaria pulcherrima (Crouan) Speg. in Michelia 1: 37. 1878. - Lachnea pulcherrima (Crouan) Gill., Champ. Fr. 76. 1880. - Scutellinia pulcherrima (Crouan) O. Kuntze, Rev. Gen. Pl. 2: 869. 1891. - Lasiobolus pulcherrimus (Crouan) Schroet., in Krypt.-Fl. Schles. (ed. Cohn) 3(2): 54. 1893. -- Cheilymenia pulcherrima (Crouan) Boud., Hist. Class, Discom, Europ. 63: 1907. — Patella pulcherrima (Crouan) Seav., N. Am. Cup-fungi (Operc.) 172. 1928. — Type: Crouan, on cow dung, Brest, Finistère, France, summer (CONC-A2388, holotype; PC-A2355, isotype).

Apothecia gregarious or in small groups, superficial, sessile, 0.5-1.0(-2) mm diam., up to 1 mm high. Receptacle at first subcylindrical, then turbinate, sometimes becoming subhemispherical, yellowish orange ('jaune orangé vif' according to Crouan, 1858); surface covered with numerous pale brown hairs; margin smooth or somewhat dentate, ending in a short hyaline raised collarette. Disc flat, even, bright orange-yellow. Hymenium up to 220 µm thick. Hypothecium scarcely differentiated. Excipulum clearly differentiated, at the margin  $35-60 \,\mu m$  wide, less distinct near the base, consisting of angular, subglobular and oblong cells  $16-40 \times 16-30 \mu m$  (textura angularis to globulosa); collarette up to 90  $\mu$ m high. Flesh rather thin, in the central part up to about 100  $\mu$ m, of rounded cells  $11-18 \mu m$  diam., intermingled with irregular hyphae 3.5-6  $\mu m$  wide. Hairs of a single type, non-rooting, arising from superficial excipular cells, single, 0-2(-3)-septate, straight, 90–200  $\mu$ m long by 11–18(–25) $\mu$ m at the base, rather brittle, gradually tapering toward the usually sharp-pointed end; the wall pale yellowish brown, rather thin (0.7-2.4  $\mu$ m). Asci cylindrical with a short stalk, rounded above,  $180-215 \times 18-22 \mu$ m, 8spored; the wall not blue with iodine. Ascospore uniseriate, ellipsoid (length/width ratio 1.5-1.9, average 1.7-1.8), at first hyaline, then pale yellowish brown, rather variable in size,  $(16.7-)18.5-24.9(-26.2) \times (10.2-)11.0-13.6(-14.5) \mu m$ , without oil-globules or granules, smooth; the outermost layer staining with methyl blue, not separating from the spore in lactic acid. Paraphyses septate, slender, cylindrical, sparsely branched, 3.5-5  $\mu$ m thick, enlarged up to 7  $\mu$ m at the tip, containing many small yellow pigment granules.

Habitat.—On old cow dung.

Specimens examined .- FRANCE: Finistère, near Brest, on old cow dung, summer, s. dat., Crouan (CONC-A2388, holotype of Ascobolus pulcherrimus; PC-A2355, isotype); Finistère, s. loc., on cow dung, 10.VI.1864, Crouan (CONC-A2374); Finistère, s. loc., s. dat. (II), Crouan (CONC-A2397).-GERMAN FEDERAL REPUBLIC, Nassau, Rheingau, near Oestrich, on cow dung, autumn, s. dat., Fuckel, in Fungi rhen. 1859 (L).

The general appearance of this species is much like that of certain species of Lasiobolus Sacc., but the mature asci are not protruding above the surface of the hymenium, the fruit bodies are not cleistohymenial, the margin ends in a collarette, the paraphyses contain a granular yellow pigment and most hairs are septate.

The present species should be placed in the genus Cheilymenia rather close to Cheilymenia insignis, both with superficial excipular hairs, ascospores with yellowish contents,

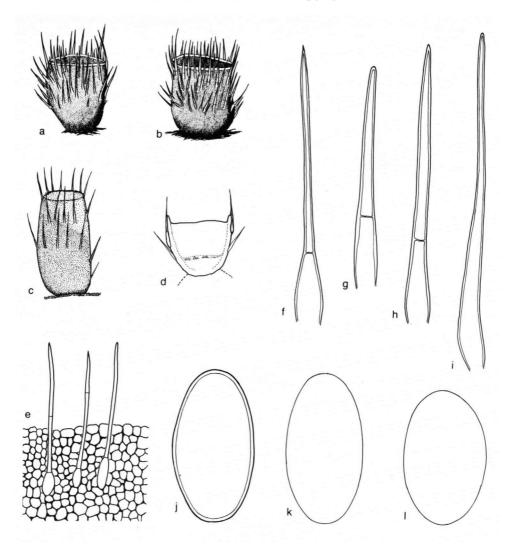


Fig. 3. Cheilymenia pulcherrima, holotype. — a, b. Habit of fruit bodies  $\times 25$ . — c. Fruit body (redrawn after original drawing in Crouan herbarium, CONC). — d. Diagrammatic section of fruit body  $\times 40$ . — e. Texture of excipulum seen from outside  $\times 160$ . — f-i. Hairs  $\times 400$ . — j. Ascospore in optical section (stained with methyl blue in lactic acid)  $\times 1600$ . — k, l. Ascospores  $\times 1600$ .

and ascospores of which the outermost layer rarely if ever loosens in lactic acid. But both species differ from each other in the colour and shape of the fruit body, the septation and shape of the hairs, the colour of the paraphyses, and the size of the ascospores.

Good coloured illustrations in the Crouan's herbarium show that the fruit bodies and the pigment of the paraphyses in *C. pulcherrima* are less vividly orange in colour than in C. insignis. In the former the colour is yellowish orange (about Munsell 2.5 Y 8/13) but in the latter deep orange (up to Munsell 4.5 YR 7/15).

No purplish hue could be found in the colour of the ascospores as mentioned by Le Gal (1961: 451).

Most descriptions of *C. pulcherrima* in literature are difficult to identify without the study of specimens. Even the exemplary description with fine illustrations by Woronin (1866) cannot be identified with certainty. As Woronin already suggests in a foot-note, his fungus is more or less intermediate in its characters between *C. pulcherrima* and *C. insignis*. Schroeter's (1893: 54) knowledge was only based on Woronin's description when he transferred *Ascobolus pulcherrimus* to *Lasiobolus*.

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