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Inocybe pusio var. *floccipes* var. nov. and some observations on the variability in this species

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Résumé – Une nouvelle variété d'*Inocybe pusio* caractérisée par la présence de caulocystides couvrant une grande partie du stipe (les 2/3 au moins) est décrite. Les caractères microscopiques de ce taxon, comme l'étude microscopique de l'holotype de *I. pusio* sont illustrées et comparées. Des observations sur la variabilité de quelques caractères macro- et micromorphologiques de *I. pusio* sont discutées.

***Inocybe* / *Inocybe pusio* / Cortinariales / taxinomie / Europe**

Abstract – A new variety of *Inocybe pusio*, characterized by the presence of caulocystidia over a great length of the stipe (-2/3) is described. Drawings of microscopical characters of this taxon, as well as a microscopical study of the holotype of *I. pusio* are added for comparison. Observations about the variability of some macro and micromorphological characters within *I. pusio* are commented.

***Inocybe* / *Inocybe pusio* / Cortinariales / taxonomy / Europe**

Resúmen – Se describe una nueva variedad de *Inocybe pusio*, caracterizada por la presencia de caulocistidios en una amplia superficie del estípite, alcanzando su tercio inferior. Se añade iconografía de sus caracteres microscópicos, así como un estudio e iconografía de los caracteres microscópicos del holótipo de *I. pusio*, para la comparación de ambos táxones. Se comenta la variabilidad de los caracteres morfológicos, tanto macro como microscópicos de esta especie.

INTRODUCTION

During the last years, both authors have gathered some collections of *Inocybe* (Fr.) Fr. determined as *Inocybe pusio* P. Karst., slightly deviating from the type by the presence of caulocystidia over a great length of the stipe. Basically all other characters of these collections agreed well with the concept of this species,

generally admitted in different monographic contributions to show caulocystidia only at the stipe apex (see Kuyper, 1986; Stangl, 1989; Bon, 1997, etc.).

Fouchier's collection (Méailles, Haute-Provence, France), attracted the attention of Bon, who made some comments about this curious form, though not validating it, assuming its possible relation to section *Splendentes*, subsection *Subbrunneae* Bon (Bon, 1999 – as *I. pusio* fo. *laticaulocystidiata* Fouchier *ad. int.*). Although no description was given, the few characters commented and the representative colour photo illustrated well the macroscopic characters of *I. pusio*, except for the more lilaceous and less violet colours of the stipe. The presence of a cortina was confirmed in our collections, placing our taxon in section *Lilacinae* R. Heim (Bon, 1997).

In addition to the references cited above, *I. pusio* has been described and/or illustrated extensively in the European mycological literature over the last century (Bataille, 1910; Lange, 1917; Heim, 1931; Alessio & Rebaudengo, 1980; Enderle & Stangl, 1981; Stangl & Veselsky, 1982; Cetto, 1983; Leisner & Kalamees, 1987; Stridvall *et al.*, 1989; Breitenbach & Kränzlin, 2000). In our opinion, however, the most helpful contributions in pointing out the variability of this species were Kühner (1955), and later Reumaux (1982) and Moënne-Loccoz *et al.* (1988), who described and/or proposed several infraspecific taxa.

MATERIAL AND METHODS

To have a more accurate vision of *I. pusio* and its variability, we compared our collections with the holotype deposited at Helsinki (H) – the latter had previously also been studied by. E. Horak, T.W. Kuyper and J. Vauras – and also with some Iberian collections labelled *I. pusio* var. *pusio* and deposited at Alcalá University Herbarium (AH).

Microscopical slides of dried material were rehydrated and prepared with 5% NH^4OH and Congo red in 1% ammonia. Spore measurements are quoted according to Heinemann & Rammeloo (1985). Drawings were made with the aid of a Zeiss drawing tube under oil immersion objective. Colours of fresh and dry fruitbodies were compared with reference colours in Munsell (1994), and the "Colour Identification Card of the Flora of British Fungi", abbreviated here (CIC/FBF), published by the Royal Botanic Gardens of Edinburgh. Terminology of cystidial elements has been adopted from Kuyper (1986). Authors abbreviations follow Kirk & Ansell (1992).

RESULTS

Inocybe pusio var. ***floccipes*** Esteve-Rav. & Fouchier, ***var. nov***. Fig. A: 1-4

= *Inocybe pusio* fo. *laticaulocystidiata* Fouchier in Bon, Bull. Féder. Assoc. Mycol. Médit. 15: 7, 1999, nom inval.

Etymology: from latin "floccus" = tuft, and "*pes*" = stipe, owing to its stipe covered by tufts of cystidia.

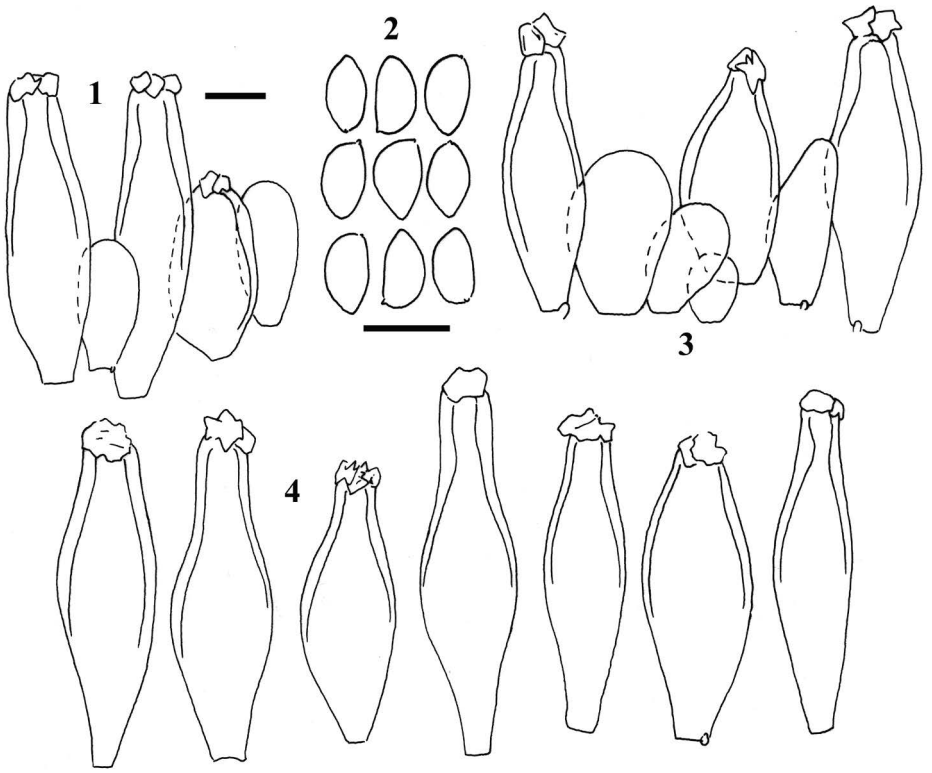

Fig. A: *Inocybe pusio* var. *floccipes* (Holotype - AH) – 1. Caulo- and paracystidia at the lower third of the stipe. 2. Spores. 3. Lamellar edge showing cheilocystidia and abundant paracystidia. 4. Pleurocystidia. Bar = 10 µm.

Latin diagnosis: *A typo differt caulocystidia et paracystidia descendentia ad basin, cheilocystidia similia. Holotypus in AH 30626.*

Description : ***Pileus*** 8-30 mm diam., when young conical-campanulate, then convex to plano-convex, often broadly and obtusely umbonate, but sometimes umbo indistinct, not hygrophanous, dark brown to brown or ochraceous-brown (7.5 YR 4/3-4/6, 3/3-3/4), sometimes mixed with a lilaceous reflection, often darker at the umbo; surface smooth, radially fibrillose outwards, at margin rimulose to distinctly rimose in adult specimens; velipellis not observed; margin inflexed when young, later hardly expanding, not striate. ***Lamellae*** moderately crowded (L = 35-55; l = 0-2), 2-5 mm broad, moderately ventricose or not, subfree, adnexed to narrowly adnate, grey-ochraceous to grey-lilaceous when young (7.5 YR 5/4, 6/4), quickly becoming ochraceous-brown to brown (10 YR 4/4-4/6, 5/4-5/6); edge uniformly whitish, sometimes concolorous, strongly fimbriate to floccose. ***Stipe*** 20-55 x 15-30 mm, cylindrical or slightly enlarged towards the base (× 4-6 mm), not bulbous, solid, lilaceous to pale grey-lilaceous (CIC-FBF nº 79) or pale amethyst-lilaceous, whitish to yellowish at the extreme base; surface distinctly

pruinose-furfuraceous all over or pruina descending to 2/3rd, and fibrillose-pruinose at the lower 1/3rd. ***Cortina*** very fugacious, only observed in very young primordia, attached very low on the stipe, absent in adult specimens, whitish. ***Context*** whitish at first, then concolorous to surface. ***Odour*** faintly spermatic.

Basidiospores (7-)7.3-*8.9*-10.5 x (4.5-)4.7-*5.35*-6.1 µm, *Q* = 1.45-*1.65*-1.87 (n = 30), amygdaliform to narrowly amygdaliform, with conical apex, smooth, thin or very slightly thick-walled (-0.4 µm), without distinct suprahilar depression, yellowish in ammonia, apiculus small. ***Basidia*** 22-32 x 7.5-10.5(-12) µm, four-spored, claviform, hyaline. ***Cheilocystidia*** (32-)40-60 x 12-20 µm, from slenderly to broadly fusiform or utriform, thick-walled, up to 1.5-2.2(-3) µm, with colourless or faintly yellowish-tinged walls, crystalliferous at apex, sometimes filled with yellowish contents in old lamellae, frequent. ***Paracystidia*** utriform, sphaeropedunculate or clavate, thin-walled, hyaline or with yellowish content, very abundant. ***Pleurocystidia*** (45-)50-65 x 14-19(-27) µm, similar in shape to cheilocystidia, thick-walled, with colourless to faintly yellowish-tinged walls, up to 1.5-2.5 µm thick, crystalliferous, rather frequent. ***Caulocystidia*** abundant over the whole stipe or descending to 2/3rd, mixed with numerous paracystidia, these elements similar in size and shape to those of the hymenium. ***Pileipellis*** a cutis composed of more or less parallel hyphae, 4-15(-23) µm, cylindrical or constricted at the septa at the subcutis, with both intraparietal and strongly encrusting brown pigment; pileus trama hardly differentiated, with colourless hyphae. ***Hymenophoral trama*** similar to pileus trama, with parallel to subparallel hyphae, cells often constricted at the septa, 3-20 µm wide, with faintly yellowish pigment; subhymenium pseudoparenchymatous, formed by 1-3 layers of subisodiametric cells. ***Clamp connections*** present at all septa.

Habitat: gregarious, in humus of continental or thermophilous *Fagaceae* forests (*Castanea sativa*, *Fagus sylvatica*, *Quercus spp*.), mainly in calcareous soils.

Material studied: **France**: Alpes de Haute-Provence: Méailles, 1400 m, 22-X-1998, under *Castanea sativa*, coll. *F. Fouchier*, F. Fouchier nº 98085 [duplo in M. Bon nº 98157]. **Portugal**: Beira Litoral: Leiria, Mata de Curvachia, 29SND 2096, 75 m, 9-XI-2000, under *Quercus faginea* in basic soil, coll. *F. Esteve-Raventós*, AH 29885. **Spain**: Barcelona: Sant Celoni, Rectoria d'Olzinelles, 31TDG 51, 200-300 m, 16-X-1991, in a mixed wood with *Quercus ilex* and *Corylus avellana*, coll. *F. Esteve-Raventós*, *J. Llistosella*, *G. Moreno* & *A. Rocabruna*, AH 24954. Burgos: Valle de Mena, Santuario de Cantonad, 30TVN 7170, 500 m, 14-X-1998, under *Fagus sylvatica*, coll. *J.M. Barrasa* & *F. Esteve-Raventós*, AH 24988. Tarragona: Montsià, Mas de Barberans, barranc de Retaule, 31TBF 6914, 1000 m, 16-X-2001, under *Fagus sylvatica*, coll. *F. Esteve-Raventós*, AH 30626 (*Holotypus*).

Additional material studied:

Inocybe pusio var. ***pusio***: **Finland**: Tavastia australis, Tammela, Syrjä, 8-VIII-1889, ad viam, coll./det. P. Karst., H -Herb. P.A. Karsten nº 1608, *holotypus*-. **Spain**: Barcelona: Premiá de Dalt, camino a Sant Bertomeu, 22-X-1999, in a mixed forest of *Pinus pinea* and *Quercus ilex*, coll. *A. Rocabruna*, AH 25337. Ciudad Real: Manzanares, Sierra de Siles, 1-V-2000, under *Populus*, coll. *F. García* & *P. Juste*, AH 26813. Huesca: Hoz de Jaca, 31-VIII-2002, in *Quercus* forest, coll. *A. González*, AH 29994. Jaén: Palomares, 17-V-1996, in *Quercus rotundifolia* forest, coll. *F. Jiménez*, AH 19399. Madrid: Rozas de Puerto Real, embalse de los Morales, 14-V-2000, under *Quercus pyrenaica*, coll. *F. Esteve-Raventós & M. Villarreal*, AH 26788. Segovia: Torreiglesias, 5-V-2003, in *Quercus rotundifolia* forest, coll. *A. Sánchez*, AH 30715.

DISCUSSION

It is unusual for a cortinate taxon in the genus *Inocybe* (Fr.) Fr. that caulocystidia and cauloparacystidia cover all or most of the stipe. Most of the cortinate Inocybe show caulocystidia, if these exist, in the upper part of the stipe (rarely descending –1/2).

I. sindonia (Fr.) P. Karst. (= *I. eutheles sensu auct. pl.*, = *I. kuehneri* Stangl & Veselsky), and probably also *I. roseipes* Malençon, are cortinate species with caulocystidia in the lower half of the stipe, but these differ from *I. pusio* var. *floccipes*, by the absence of typical paracystidia in between the cystidia. A possible ontogenetic explanation of the presence of basal caulocystidia in *I. sindonia* was commented by Kuyper (1986: 7), who considers this case as "exceptional".

Among the numerous specimens examined in the five collections of *I. pusio* var. *floccipes*, only one specimen showed a distinct cortina attached close to the stipe base. Such a position of the partial veil in young primordia can explain the presence of a caulohymenium (cystidia and paracystidia) in this area of the stipe. This cannot be considered a casual phenomenon, as it has been observed in five different collections from different countries, and was mentioned for the first time by Kühner (1955), referring to "form 2º" in his comments on *I. pusio*. The other morphological characters of var. *floccipes*, however, do not allow to make a clear distinction from var. *pusio*. It is worth mentioning that two other features have revealed to be slightly deviating, i. e. the colour of the stipe, which never shows clear violaceous tinges, and the slightly thicker cystidia walls.

The variable distribution of caulocystidia in *I. pusio* was already reflected in Kuyper's key (1986) where this species can be identified either by choosing caulocystidia only present at the insertion of the stipe (extreme apical part, -1/10 or 1/6), or by choosing present at a lower position (-1/3). This variation (first noted by Kühner, 1955), has been studied and described by Reumaux (1982) and Moënne-Loccoz *et al*. (1988), who proposed some different infraspecific taxa in *I. pusio*, *i. e*. fo. *velata* Reumaux, fo. *elegans* Reumaux and fo. *salmoncipes ad int*.

I. pusio fo. *velata* shows a well-developed velipellis, which causes, in the course of development, the presence of arachnoid or floccose whitish patches on the pileus. This form was described as having very few, dispersed caulocystidia, mixed with caulocystidioid hairs only at the extreme apex of the stipe.

I pusio fo *elegans* has an excoriate pileus and (sub)capitate, thick-walled cystidia – a type of cystidia that, as Kuyper (1986) has emphasized, seems to be very characteristic for *I. griseolilacina* J.E. Lange, with which he considers this form to be conspecific.

According to Moënne-Loccoz *et al*. (1988), *I pusio* fo *salmoncipes* shows neither violet nor lilaceous tinges on the stipe and may represent *I. huijsmanii* Kuyper, but a comparison between the illustrations of Moënne-Loccoz (Pl. 254) and those of Stangl (1989: Tafel 12/7) for this species, reveal clear macroscopical differences. We have neither found these two taxa in the field nor studied herbarium material.

The interpretation of *Inocybe personata* Kühner by some French authors (e.g. Bon, 1997; Moënne-Loccoz *et al.*, 1988), remains doubtful to us. Having studied several collections determined as *I. personata* by Kühner himself (from G, lectotype not yet selected), Esteve-Raventós & Villarreal (2001) concluded that this species is a synonym of *I. griseolilacina*. This interpretation is very similar to that of Kühner in Kühner & Romagnesi (1953: 511), who considered it a variety of Lange's species, having more reddish tinges in the context of the upper part of the

stipe. The other characters of *I. personata* (see description in Kühner, 1955) do not differ from those of *I. griseolilacina*. Having studied numerous collections of the latter common species, we think that the colour of the upper stipe context is variable : some collections having more reddish tinges, while others have only pure lilaceous tinges. In our opinion, the most helpful character to recognize this species is the presence (in variable number) of (sub)capitate cystidia. The pelargonium odour is sometimes considered significant to recognize *I. personata*. As it is the case of many species of *Inocybe*, however, the odour can be noted and/or described in many different ways, and seems easily influenced by environmental factors, time and/or personal perception, as also suggested by Kühner (*loc. cit.*) in his description of the odour for *I. personata* "passant facilement pour vague, légère ou spermatique, ..les fragments découpés pour le préparation de l'herbier dégagent une odeur acidulée subpélargoniée, ...".

Morphological variability of *Inocybe pusio* in the Iberian Peninsula

We compared the holotype of *I. pusio* (Fig. B: 1-4), deposited at (H) with several collections from the Iberian Peninsula. All collections, determined as var. *pusio,* show caulocystidia in the upper 1/3rd or near the insertion area of the stipe and have clear violet-lilaceous tinges in the stipe and, in some cases, also in the lamellae and/or on the pileus surface. Some collections with a well developed velipellis would correspond to forma *velata* Reumaux.

Collections showing caulocystidia mixed with paracystidia descending to 2/3rd or all over the stipe, have been named var. *floccipes*, these also show slight chromatic variations on the stipe.

I. pusio is macroscopically distinghished by the smooth pileus, which becomes typically rimose towards the margin with age, and by the presence of violaceous colours in the stipe and of fusiform to utriform cystidia with walls not distinctly yellowing with ammonia.

In the type collection, caulocystidia are present descending to the upper 1/3rd of the stipe, and are mostly broadly fusiform to broadly utriform, often broader than 20 µm. However, cystidia diameter has been observed as very variable in most of the collections studied. Variability has been observed also in the following characters:

Size and habit of basidiomata vary greatly as already remarked by Reumaux (1982) with slender and robust forms encountered within the same collection, growing often gregarious, but more rarely fasciculate.

Aspect of pileus and pileipellis: mostly depending on the velipellis presence, which exerts a great influence on the degree of excoriation of the pileipellis (see Kuyper, 1986: 5), which may exceptionally results in a subsquamulose pileus centre.

Presence and distribution of violet or lilaceous colours: these are always present on the stipe, but collections with pale violaceous colours on the lamellae or similar tinges or reflections on the pileus are not uncommon. These colours, however, may vanish with age and become ochraceous to yellowish, depending on environmental factors.

Spores: though typically amygdaliform with (sub)acute apex, these may appear slightly papillated at the apex, *i. e.* subcitriform, in some collections ; the latter type always being related to heterosporic collections with presence of 2-spored basidia and anomalous, sometimes gigantic, spores. Heterosporic populations are often encountered in this species.

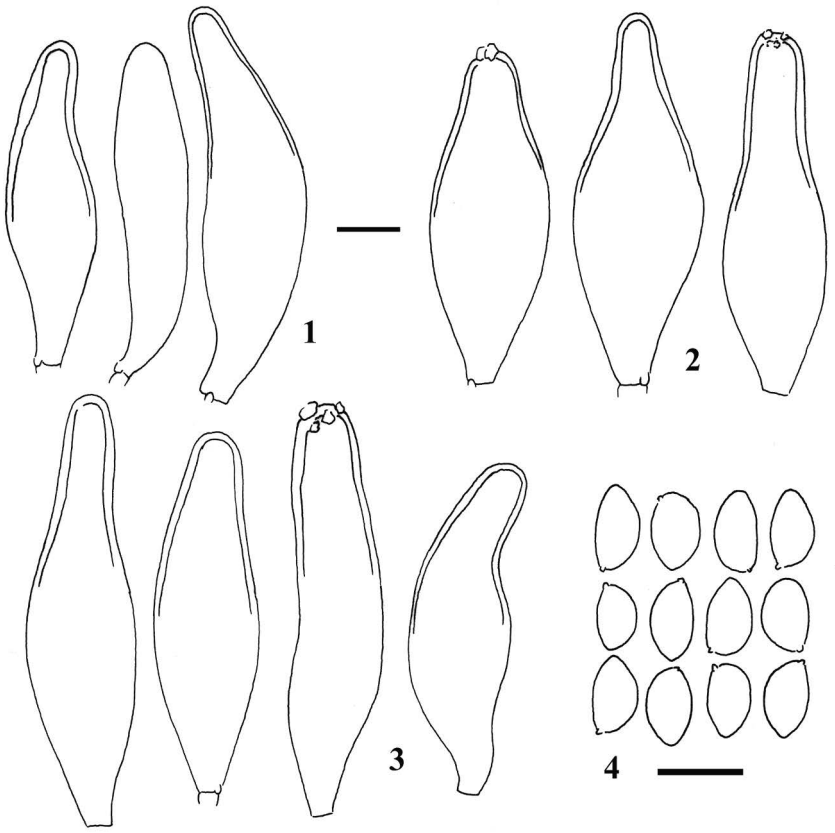

Fig. B: *Inocybe pusio* (Holotype — H) – 1. Some caulocystidia at the upper third of the stipe. 2. Cheilocystidia. 3. Pleurocystidia. 4. Spores. Bar = 10 µm.

Cystidia broadly to narrowly fusiform or utriform – the frequency depending on collections – crystalliferous at apex, with a slightly thickened wall (0.8-2 µm), colourless or faintly yellowish in ammonia; content normally colourless (in old specimens some yellowish intracellular pigment may be present, especially in cheilocystidia).

Paracystidia extraordinarily frequent at the lamellae edge, mixed with abundant cheilocystidia and rendering the edge sterile; very abundant paracystidia mixed with caulocystidia in var. *floccipes*. In var. *pusio* (fo. *pusio* and fo. *velata*) practically absent or present only in a very narrow zone at the apex of the stipe, mixed with caulocystidia and caulocystidioid hairs.

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