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Taxonomy of *Alnus*-associated hypogeous species of *Alpova* and *Melanogaster* (Basidiomycota, Paxillaceae) in Europe

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Abstract – Morphological analysis of basidiomata of gastroid Boletales (*Alpova* and *Melanogaster*) found in relation with *Alnus* trees in France revealed the existence of five distinct *Alnus*-associated species: *Alpova alpestris* sp. nov., *A. corsicus* sp. nov., *Melanogaster luteus* Zeller, *M. rivularis* sp. nov., and an unnamed species (*Alpova* sp. related to the North American *A. cinnamomeus*). The genus *Alpova* is here restricted to its original sense, as *Alnus*-associated species with small spores and pseudoparenchymatic subpellis, and the presence of "buffer cells" in gleba. Phylogenetic position, host specificity and geographical distribution of each species are summarized.

Taxonomy / *Alpova diplophloeus* / Boletales / Paxillineae

INTRODUCTION

In traditional systematics gastroid Basidiomycota with gelatinized gleba are usually classified in the old genus *Melanogaster* Corda (Corda, 1831). In 1931, Dodge described a new genus *Alpova* C.W. Dodge to accommodate a new species *A. cinnamomeus* C.W. Dodge; this species collected in Royal Isle, Michigan, under alders (likely *Alnus incana* subsp. *rugosa* (Du Roi) R.T. Clausen), was characterized by a cellular peridial structure. This pseudoparenchymatous structure was described by Zeller (1947) who noted the presence of characteristic "buffer cells" in the gelatinized lacunae of the gleba. *Alpova cinnamomeus* was then only cited in relation to *Alnus* trees, but its specificity was only established by Trappe (1975) who synonymized it with the formerly described *Melanogaster diplophloeus* Zeller & C.W. Dodge. At the same time Trappe (1975) extended the definition of the genus *Alpova* to all pale-spored species with more or less gelatinized gleba, formerly described in the genera *Melanogaster* and *Rhizopogon*. This emendation has been widely accepted in modern literature, e.g. by Beaton *et al.* (1985) and Liu *et al.* (1990) who integrated in *Alpova* several new Asian and

Australian pale-spored species, and by Gross (1980), Jülich (1984), and Montecchi and Sarasini (2000) who recognized four European species of *Alpova*: *A. diplophloeus, A. klikae* (Mattir.) Trappe, *A. microsporus* (Velen.) Trappe, and *A. rubescens* (Vitt.) Trappe.

However, molecular studies of the *Melanogaster-Alpova* group have indicated that *Alpova* sensu Trappe (1975) is polyphyletic, and confirmed the necessity to redefine the genus *Melanogaster* (Grubisha *et al.*, 2001; Jarosh and Besl, 2001; Bougher and Lebel, 2002; Binder and Hibbett, 2006; Halász, 2008). Recently, Nouhra *et al.* (2005) and Vizzini *et al.* (2010) proposed to come back to the original definition of *Alpova*, as strictly *Alnus*-associated species. In summary, only two species of *Alpova* are now fully accepted by taxonomists: *A. diplophloeus* described from North America, and *A. austroalnicola* L.S. Domínguez from South America (Nouhra *et al.*, 2005); no specific name is available at this date for any well-defined European species, usually mentioned as *A. diplophloeus* or *Melanogaster microsporus* in the literature (see Table 1). Moreover DNA sequences available in Genbank under the name *Alpova diplophloeus* clearly show that several taxa are confused under this name (Rochet *et al.*, 2011).

In the context of a large-scale study of fungal populations associated with *Alnus* spp. in France, we have collected basidiomata of *Alnus*-associated ectomycorrhizal species in various localities in France (especially Alps, Corsica, Pyrénées, Massif Central, and Nord-Pas-de-Calais). We propose here a revision of European hypogeous *Alnus*-associated Boletales based on our collections of *Alpova* and *Melanogaster* and other specimens sent by our correspondents from Montenegro and Romania. We aim to define the taxonomy of European species of *Alpova* and *Melanogaster*, as a prelude to a more detailed paper about the systematics and evolution of these taxa based on a multigene phylogeny.

MATERIAL AND METHODS

Basidiomata sampling – Sporocarps were collected in various habitats in the French Alps (under *Alnus incana* and *A. alnobetula* subsp. *alnobetula*) and Corsica (under *Alnus cordata*, *A. glutinosa*, and *A. alnobetula s*ubsp. *suaveolens*); no sporocarp have been observed under *Alnus glutinosa* outside Corsica, despite careful investigations. When possible, fresh specimens were were photographed and described after sampling; all specimens were cut and air-dried. Specimens are preserved in the Herbarium of the Faculté des Sciences Pharmaceutiques et Biologiques, Lille (LIP); isotypes are deposited at the herbarium of the Eidgenössische Technische Hochschule, Zürich (ZT).

Observations – Microscopic characters were observed with an light microscope Nachet at × 100, × 400 and × 1000 magnifications on hand-sectioned mounts in Melzer's reagent, 10% KOH, and Congo red 1% in 10% NH_4OH after rehydrating in 10% KOH for a few minutes. Peridium structure is described from both radial cuts and tangential cuts (scalps). Descriptive terminology is taken from Zeller (1939). For all collections, at least 30 spores have been measured (see Table 2), statistical treatments and notations follow Fannechère (2005); measurements were made on the software Mycomètre 2.02 (Fannechère, 2009). In the following descriptions spore dimensions are given as follows: (minimum value) 1^{st} decile – ***average value*** – 9^{th} decile (maximum value), calculated on all spores measured for each species (for details see tab. 1).

Table 1. Reports and revision of *Alpova* and *Melanogaster* citations in association with *Alnus* in European literature

Reference	*Species of* Alnus	*Area*	*Name used in publication*	*Revised name (see below)*
Favre (1960: 585)	*A. alnobetula* subsp. *alnobetula*	Switzerland (Graubunden)	*Melanogaster microsporus* Velen.	*Alpova alpestris*
Clémençon (1977)	*A. alnobetula* subsp. *alnobetula*	Switzerland (Vaud, Graubünden)	*Alpova diplophloeus* f. *europaeus*	*Alpova alpestris*
Schmid-Heckel (1985)	*A. alnobetula* subsp. *alnobetula*	Germany (Bavaria)	*Alpova diplophloeus* and *A. klikae*	*Alpova alpestris*
Kers (1986)	*A. incana*	Norway (Oslo)	*Alpova diplophloeus*	*Melanogaster luteus*
Montecchi & Lazzari (1989)	Not precised (*A. glutinosa* o *A. incana*)	Italy (Appenins)	*Alpova diplophloeus*	*Melanogaster luteus*
Eckblad & Lange (1997: 295)	Not precised	Norway, Sweden	*Melanogaster microsporus* Velen.	*Melanogaster luteus*
Gross (1980), Gross *et al.* (1980)	*A. alnobetula* subsp. *alnobetula*	Germany (Bayern)	*Alpova diplophloeus* f. *diplophloeus*	*Alpova alpestris*
Montecchi & Sarasini (2000)	*A. glutinosa* and *A. incana*	Italy, Switzerland (Ticino)	*Alpova diplophloeus*	*Melanogaster luteus*
Franchi & Marchetti (2001)	*A. alnobetula* subsp. *alnobetula*	Italy	*Alpova diplophloeus*	*Alpova alpestris*
Moreau (2003)	*A. alnobetula* subsp. *alnobetula*	France (Savoie)	*Alpova diplophloeus* subsp. *europaeus*	*Alpova alpestris*
Jamoni (2008: 139)	*A. alnobetula* subsp. *alnobetula*	Italy (Monte Rosa)	*Alpova diplophloeus*	*Alpova alpestris*

DNA extraction, PCR amplification and sequencing – Sporocarps were collected from multiple stands of the four European alder species at various locations in France. Tissue samples (part of the gleba) were fixed in a CTAB 2X Buffer (100 mM Tris–HCl (pH 8), 1.4 M NaCl, 20 mM Na2 EDTA, 2% CTAB) until DNA extraction. The DNA regions and primers used for PCR are: the nuclear rDNA ITS using the primer set ITS-1F/ITS-4B (Gardes & Bruns 1993). Other genes were investigated, with congruent results with ITS only (not shown, see Rochet *et al.* 2011): the rpb2 gene using bRPB2-5F/bRPB2-7.1R (Matheny *et al.* 2005), the gpd gene using GPD-F/GPD-RA (Johannesson *et al.* 2000, modified in Jargeat *et al.* 2010), and the hyper variable domain V9 of the mitochondrial SSU-rDNA using the V9U/V9R (Gonzalez *et al.* 1998; Mouhamadou *et al.* 2008). Amplifications were carried out in 25 µl reaction containing 0,2 µl of GoTaq polymerase of 5 U/µl (Promega), 5 µl of Buffer 5X (Promega), 0.25 µl of 5 µM for each primer, 5 µl of dNTPs 5mM, 2 µl of template DNA and 15.8 µl of nuclease free water. Standard cycling parameters were an initial denaturation step at 94°C for 3 minutes, 35 cycles of denaturation at 94°C for 45 secondes, annealing for 45 secondes at different temperatures depending on the primer set (50° for rpb2, 53° for V9, and 55°C for ITS and gpd), and elongation at 72°C for 1 min, followed by a final elongation step at 72°C for 10 min. Post-cycling, samples were held at 4°C. Unsuccessfully amplified samples were subjected to multiple amplifications at various DNA concentrations. Sequencing was done by MilleGen (Labège, France).

Alignment and phylogenetic analysis – ITS sequences were manually edited using Sequencher 4.8. Gene Codes (Ann Arbor, MI). They were aligned using MAFFT version6 using the LINS-i method with standard settings (Katoh *et al.* 2005) and subsequently carefully refined by eye. Phylogenetic analyses were conducted on sequence dataset using Bayesian inference as implemented in MrBayes 3.1.2 (Huelsenbeck & Ronquist 2001).

Bayesian inference – Modeltest 3.7 (Posada & Crandall 1998) was run to select a model of sequence evolution for the ITS dataset. Appropriate DNA substitution models and its parameters were determined based on the comparisons of negative log likelihood values in Modeltest 3.7 (Posada & Crandall 2001) implemented in PAUP*4.0b10 (Swofford 2001). Likelihood and prior settings were changed to meet with the settings corresponding to the model found. The analyses were initiated with random starting trees and one cold and three incrementally heated Markov chain Monte Carlo (MCMC) chains were run for 5,000,000 generations. Trees were sampled every 1000 generations. MCMC runs were repeated twice. Stationarity of the Markov chain was ascertained by plotting likelihood values against number of generations for apparent stationarity. The first 1000 to 2000 trees were discarded as burn-in, and the remaining trees were used to calculate a 50% majority rule tree and to determine the posterior probabilities for the individual branches.

TAXONOMY AND SYSTEMATICS

Taxonomy of *Melanogaster microsporus*

As a coincidence, the two earliest reports of presumed *Alpova* species in Europe were described as new species under the same name *Melanogaster microsporus*, by Velenovský (1922), then by Mattirolo (1931). As pointed out by Zeller (1939) the identity of *M. microsporus* Velen. is a key question in taxonomy of small-spored gastroid *Paxillaceae*. It can be asked whether both names are homonyms *and* synonyms, as it was suggested by Söhner and Knapp (1947: 154) and Svrček (1958: 545), and if any of these names could be applied to our collections.

Melanogaster microsporus Velen. (Velenovský 1922) was placed by Trappe (1975) in the genus *Alpova* on the basis of Knapp's (1954b) and Svrček's (1958) descriptions. In fact the type collection was revised by Knapp (1954b: 150), who only briefly described its spores ("gelbbraune, reif"[1]): his full description is based on a later collection from Arosa (Swiss Alps, leg. E. Rahm), with a gleba "jung zitronengelb-gelbgrünlich[2]" in contradiction to the protolog. Our revision of the holotype collection (coll. J. Velenovský; PRM) clearly shows yellow-brown, thick-walled spores, measuring 5.6-*6.51*-7.5 × 3.1-*3.71*-4.2 µm (see Table 2), mainly ellipsoidal with remnants of sterigmata and a typical *Melanogaster*-type peridium. In our opinion this specimen is not distinct from *Melanogaster broomeanus* Berk.; no clue indicates that Velenovský's collection was associated with any species of *Alnus* (J. Holec, *pers. com.*).

1. Yellow-brown, mature.
2. Young lemon yellow, greenish yellow.

Table 2. Estimated spore dimensions of European studied collections. 1st and 9th deciles (D1,9) and average value (bold, italic) are given according to Fannechère (2005, 2009). *: South-American collection (leg. E. Nouhra). **: Type collection of *M. microsporus* Velen. (PRM), excluded from *Alpova* (see Excluded taxa below).

Voucher	*Number of measures*	*Spore length (µm)*	*Spore width (µm)*	*Quotient*	*Volume (µm³)*
Alpova alpestris (Alps) 07082629-S053 (typus)	85	4.4-***4.99***-5.6	2.2-***2.46***-2.8	1.82-***2.04***-2.27	11.1-***16.06***-22.3
Alpova alpestris (Alps) 95081604	120	4.7-***5.53***-6.9	2.4-***2.71***-3.2	1.79-***2.05***-2.28	14.7-***22.09***-37.5
Alpova alpestris (Alps) 07082632	120	4.9-***5.57***-6.3	2.3-***2.59***-2.8	1.89-***2.16***-2.45	14.8-***19.81***-25.7
Alpova alpestris (Alps) 09082202	120	3.9-***5.11***-6.0	1.9-***2.38***-2.7	1.89-***2.15***-2.38	8.2-***15.84***-22.9
Alpova alpestris (Alps) 09082802	60	5.0-***5.61***-6.1	2.3-***2.47***-2.7	2.03-***2.27***-2.51	14.4-***18.12***-22.2
Alpova alpestris (Alps) 09082300	60	4.9-***5.43***-6.0	2.5-***2.77***-3.1	1.74-***1.97***-2.25	16.6-***22.02***-28.1
Alpova alpestris (Corsica) 08090302-S159 (typus)	125	4.4-***5.08***-5.6	2.2-***2.48***-2.8	1.81-***2.06***-2.34	11.9-***16.5***-22.1
Alpova alpestris (Corsica) 04101113-S60	120	4.8-***5.52***-6.2	2.3-***2.48***-2.7	1.92-***2.23***-2.52	13.8-***17.92***-22.6
Alpova alpestris (Romania) 09082300	120	4.9-***5.39***-5.9	2.4-***2.69***-3.0	1.79-***2.02***-2.26	15.5-***20.61***-26.5
Alpova corsicus 05091024	120	4.5-***4.26***-5.0	1.9-***2.19***-2.4	1.86-***2.10***-2.3	8.8-***11.58***-14.2
Alpova corsicus 04101021 (typus)	120	4.5-***5.01***-5.6	1.9-***2.15***-2.5	2.00-***2.35***-2.76	8.5-***12.36***-17.5
Alpova corsicus 04101021bis	120	4.0-***4.57***-5.1	1.8-***2.07***-2.3	1.91-***2.22***-2.54	7.6-***10.37***-13.2
Alpova corsicus 05102601	120	4.3-***4.73***-5.1	1.9-***2.15***-2.4	1.95-***2.21***-2.50	8.4-***11.61***-14.7
Alpova corsicus 04101114	68	4.2-***4.72***-5.3	2.14-***2.29***-2.5	1.85-***2.07***-2.36	10.6-***13.06***-16.19
Alpova corsicus (long-spored) 07090813	120	5.5-***6.07***-6.6	2.5-***2.75***-3.0	2.01-***2.22***-2.43	18.0-***24.31***-29.8
Alpova cf. *cinnamomeus* 09082702	120	4.4-***4.93***-5.5	2.1-***2.33***-2.5	1.83-***2.13***-2.49	10.9-***14.14***-17.4
**Alpova austroalnicola* L.S.Dominguez 2290 (paratype)	100	4.8-***5.33***-5.9	2.2-***2.47***-2.76	1.93-***2.17***-2.41	12.3-17.27-23.0
Melanogaster luteus 09082801	120	5.3-***6.01***-6.8	2.4-***2.75***-3.1	1.95-***2.20***-2.51	16.8-***24.17***-31.7
Melanogaster luteus Peric 30.VII.2006	120	5.3-***5.90***-6.5	2.8-***3.08***-3.3	1.75-***1.92***-2.12	23.2-***29.53***-37.2
Melanogaster rivularis 08090514-S191 (type)	120	5.1-***5.65***-6.2	2.7-***2.91***-3.1	1.74-***1.95***-2.17	20.5-***25.25***-31.0
***Melanogaster microsporus* (type)	57	5.6-***6.51***-7.5	3.1-***3.71***-4.2	1.53-***1.76***-2.03	30.3-***48.30***-67.2

Melanogaster microsporus Mattir. was originally described from Europe (Italy, province of Como, around Rodero) from a mixed sloping forest of *Castanea* and *Pinus sylvestris* (Mattirolo 1931). Although Mattirolo does not mention *Alnus* as a possibly associated tree, the presence of *Alnus glutinosa* sparsely intermixed with *Castanea* and *Pinus* in this area has been confirmed by the Italian mycologist Mario Cervini (*pers. com.*). If *M. microsporus* Mattir. can be stated as an *Alnus glutinosa* (or *A. incana*)-associated taxon, then it is a very rare species, considering the very few reports of *Alpova* and *Melanogaster* under these trees (Kers 1986; Montecchi and Lazzari 1989). No original collection could be traced in ZT and TO (where most of Mattirolo's collections are preserved), but we could study two recent collections, from Monte Negro and the French Alps, both under *Alnus incana*, and confirm its characters as they were well described by Mattirolo (especially golden yellow peridium and small size), and its phylogenetic position. It must be named *Melanogaster luteus* Zeller, a new name replacing Mattirolo's illegitimate name and based on the same type (Zeller 1939; Fogel 1977). It is redescribed and illustrated by Perič and Moreau (2010).

The hypothesis of Knapp (1954b), Svrček (1958) and Montecchi and Lazzari (1989), of a synonymy between *Melanogaster microsporus* Mattir. and *M. microsporus* Velen., is rejected here, as well as their position in the genus *Alpova* suggested by Trappe (1975).

Revision of *Alnus*-associated European collections

The European collections at our disposal could be split morphologically into two distinct groups. The first, with pseudoparenchymatous peridial structure, olivaceous gleba before maturity, presence of buffer cells in gleba, and small spores yellowish *sub micr.*, strictly conform to the definition of the genus *Alpova* in its restricted sense (Dodge, 1931; Nouhra *et al.*, 2005). The second, with prosenchymatous peridial structure, brown to blackish gleba even when young, greyish to dark spores with longer remnants of sterigmata and absence of buffer cells, match the definition of the genus *Melanogaster*. The authors checked in vain for the presence of buffer-cells (Zeller 1947; Nouhra *et al.*, 2005) in the gleba of all studied species of *Alpova* and *Melanogaster*, but it is impossible to affirm their absence in most species because this character is never mentioned in the literature.

Considering only *Alpova*, three distinct European species are characterized by micromorphological characters. They are also resolved by phylogenetic analysis based on ITS sequences, as well as by multigene phylogenies (ribosomal nuclear DNA: RBP2; protein-coding gene: GPD; mitochondrial gene: V9; data not shown). When ITS sequences from North American samples (archived in Genbank as "*A. diplophloeus*") are added to the analysis, all represent allied but distinct clades (data not shown).

One of our species (*A. corsicus*) is apparently endemic to Corsica and appears under all species of *Alnus*, one (*A. alpestris*) was found widespread in the Alps, the Carpathians and Corsica, always under *Alnus alnobetula*; another unnamed species (*A. sp.*), morphologically and ecologically the closest to the original description of *A. cinnamomeus* (Dodge 1931), is known by a single specimen found under *Alnus alnobetula* in the Alps. Morphological characters are few but characterize the species well enough to make them recognizable by mycologists (see Key below), particularly spore width and shape, outer peridium structure, development of rhizomorphs, and basidium shape prove to be very

useful characters. Gleba structure, rounded *versus* daedalean lacunae, and oxydation when cut, seem to differenciate *A. alpestris*/*A.* aff. *cinnamomeus* from *A. corsicus*. Macroscopically, aspect of peridium, odour, and intensity of staining require more observations to be confirmed as good field characters.

The two *Alnus*-associated species of *Melanogaster*, one in continental Europe (*M. luteus*), the other in Corsica (*M. rivularis*), are easily separable from *Alpova* in the field by their dark gleba colour when mature, their unchanging context when cut and microscopically also by their plectenchymatous peridium (subpellis). Both species differ also in size (*M. rivularis* is 2-4 times more voluminous than *M. luteus*) and in color (*M. luteus* has a bright yellow surface).

Molecular phylogeny

Each selected gene (ITS, GPD, RPB2, V9) has been analyzed individually and after concatenation. All results obtained were highly congruent regarding species delimitation and phylogenetic arrangement. ITS sequences were easily aligned and sufficient to get a strongly supported phylogeny for the whole *Alpova* + *Melanogaster* lineage, with a good resolution at species level. For the purpose of this paper only a partial ITS phylogeny (Fig. 7) is shown, a multigene analysis restricted to species of *Alpova* has already been presented by Rochet *et al.* (2011).

The presented phylogeny (Fig. 7) especially illustrates the position of the two *Alnus*-associated species *Melanogaster luteus* (continental Europe) and *M. rivularis* (Corsica) in the clade /Melanogaster. As far as European species are considered, the genus *Melanogaster* is clearly split into three lineages, corresponding respectively to sect. *Melanogaster*, *Variegati*, and *Rivulares* (see Descriptions below). The North American species *'Alpova' trappei* Fogel clearly belongs to this genus but remains unresolved between sect. *Melanogaster* and the *Variegati*+*Rivulares* clade. Other extra-European species of *Melanogaster* were not included in this analysis due to the absence of available sequences in public databases.

The genus *Alpova* is confirmed as a sister group of *Melanogaster* (Nouhra et al. 2005), and includes so far only *Alnus*-associated species with buffer cells in their gleba. Their analysis is already treated by Rochet *et al.* (2011).

Descriptions

In the present study, we accept the most restrictive definition of *Alpova*, following Nouhra *et al.* (2005) and Vizzini *et al.* (2010), summarized by the following characters: basidiomata globose to subglobose, hypogeous; gleba constituted by gelatinized lacunae without hymenial structure (lack of hymenial palisade when mature); spores smooth, slightly thick-walled, generally biguttulate; basidia small, generally eight-spored; clamps always present; peridium two-layered, peridiopellis filamentous with or without rhizomorphs or dermatocystidia, subpellis pseudoparenchymatous; buffer cells persistent in mature gleba (see Fig. 3H); forming ectomycorrhizae with *Alnus* species. Species with prosenchymatous peridial structure are maintained in the genus *Melanogaster*.

I. *Alpova* C.W. Dodge, *Ann. Missouri Bot. Gard.* 18: 461. 1931.

= *Melanogaster* sect. *Microspori* Svrček, *Flora ČSR* B(1): 545. 1958 (inval., no latin diagnosis).

Table 3. References of collections used in phylogenetic analysis (see Fig. 7).

Alpova alpestris P.-A. Moreau & F. Richard	PAM07082724	F	Savoie	Bourg-Saint-Maurice	Aalnobet	HQ714684
Alpova alpestris P.-A. Moreau & F. Richard	PAM07082632	F	Savoie	Landry	Aalnobet	HQ714686
Alpova alpestris P.-A. Moreau & F. Richard	PAM07090501	F	Haute-Corse	Casamacchioli	Asuav	HQ714691
Alpova alpestris P.-A. Moreau & F. Richard	PAM07090501	F	Haute-Corse	Casamacchioli	Asuav	HQ714692
Alpova alpestris P.-A. Moreau & F. Richard	PAM07082629	F	Savoie	Bourg-Saint-Maurice	Aalnobet	HQ714696
Alpova alpestris P.-A. Moreau & F. Richard	PAM07082632	F	Savoie	Landry	Aalnobet	HQ714697
Alpova alpestris P.-A. Moreau & F. Richard	PAM08082716	F	Savoie	Crest-Voland	Aalnobet	HQ714711
Alpova alpestris P.-A. Moreau & F. Richard	PAM08090302	F	Corse du Sud	Bastelica	Asuav	HQ714721
Alpova alpestris P.-A. Moreau & F. Richard	PAM08082103	F	Savoie	Bourg-Saint-Maurice	Aalnobet	HQ714772
Alpova alpestris P.-A. Moreau & F. Richard	EDB080829	F	Savoie	Bourg-Saint-Maurice	Aalnobet	HQ714775
Alpova alpestris P.-A. Moreau & F. Richard	PAM09082802	F	Savoie	Crest-Voland	Aalnobet	HQ714776
Alpova alpestris P.-A. Moreau & F. Richard	PAM09082201	F	Savoie	Crest-Voland	Aalnobet	HQ714777
Alpova alpestris P.-A. Moreau & F. Richard	PAM09082202	F	Savoie	Crest-Voland	Aalnobet	HQ714778
Alpova austroalnicola L.S. Domínguez	LS Dominguez 2290	ARG	Salta province	Sancta Victoria, Los Toldos	Aacum	HQ714793
Alpova cf. cinnamomeus C.W. Dodge	PAM09082702	F	Savoie	Beaufort	Aalnobet	HQ714779
Alpova corsicus P.-A. Moreau & F. Richard	PAM04101021B	F	Haute-Corse	Ponte Leccia	Aglut	
Alpova corsicus P.-A. Moreau & F. Richard	PAM05102601	F	Haute-Corse	Fort Stella	Acord	HQ714698
Alpova corsicus P.-A. Moreau & F. Richard	PAM07090803	F	Corse du Sud	Bastelica	Asuav	HQ714699
Alpova corsicus P.-A. Moreau & F. Richard	FR2	F	Haute-Corse	Omita	Acord	HQ714766
Alpova corsicus P.-A. Moreau & F. Richard	FR27	F	Haute-Corse	Ponte Leccia	Aglut	HQ714768
Alpova corsicus P.-A. Moreau & F. Richard	FR28	F	Haute-Corse	Ponte Leccia	Aglut	HQ714769
Alpova corsicus P.-A. Moreau & F. Richard	FR29	F	Haute-Corse	Ponte Leccia	Aglut	HQ714770
Alpova 'diplophloeus' (Zeller & C.W. Dodge) Trappe	UBC F15185	CAN	British Columbia	Capilano Riv. Reg. Park	Arub?	DQ384577

Table 3. References of collections used in phylogenetic analysis (see Fig. 7). *(continued)*

Alpova 'diplophloeus' (Zeller & C.W. Dodge) Trappe	3824.0	USA	Oregon	Lincoln Co., Cape Perpetua	Arub	DQ989496
Alpova 'diplophloeus' (Zeller & C.W. Dodge) Trappe	OSC59767	SW	Dalarna		Ainc	DQ989497
Alpova 'diplophloeus' (Zeller & C.W. Dodge) Trappe	PK2102	CAN	British Columbia	Malcolm Knapp Research Forest	Arub	DQ989498
"Alpova" trappei Fogel *(1)*		USA				AF074920
Melanogaster ambiguus (Vittad.) Tul. & C. Tul.	B-2409	HU		Szilvasvarad		AJ555514
Melanogaster broomeanus Berk.	B-2331	HU		Szelcepuszta		AJ555530
Melanogaster luteus Zeller	PAM09082801	F	Savoie	Hauteluce	Ainc	HQ714780
Melanogaster luteus Zeller	Mon06	MTN		Opasanica	Ainc	HQ714794
Melanogaster macrosporus Velen.	B-1438	HU		Algyo		AJ555523
Melanogaster rivularis P.-A. Moreau & F. Richard	PAM08090514	F	Haute-Corse	Asco	Aglut	HQ714731
Melanogaster rivularis P.-A. Moreau & F. Richard	PAM08090514	F	Haute-Corse	Asco	Aglut	HQ714767
Melanogaster tuberiformis Corda	B-1295	HU		Sfantu Gheorghe		AJ555527
Melanogaster variegatus (Vittad.) Tul. & C. Tul.	B-1688	HU		Gant		AJ555524
Melanogaster vittadinii Soehner & Knapp	33090	HU		Pilisszentkereszt		AJ555525

I.1. ***Alpova alpestris*** P.-A. Moreau & F. Richard, *sp. nov.* Fig. 1A-C, 2A-H, 5E-J

Mycobank MB 518391

Etymology: from the Alps, where the species is especially common and widespread under *Alnus alnobetula*.

= *Melanogaster microsporus* Velen. sensu Knapp (1954a, 1954b) *pro parte*, sensu Favre (1960)

= *Alpova diplophloeus* sensu auct. europ.

= *Alpova diplophloeus* f. *europaeus* sensu Clémençon (1977)

= *Alpova diplophoeus* f. *diplophloeus* sensu Gross (1980)

Illustrated references: Wiedmer *et al.* (2001), as *A. diplophloeus* (mycorrhizae); Jamoni (2008: 139), as *A. diplophloeus*; Buyck (2009: 14), as *A. diplophloeus*.

Basidiomata 0.8-2.5 cm in longitudine, 0.8-1.5 cm in diametro, a globosis ad oblonga, rarius irregulariter extensa vel constricta, basi haud vel paene distincta; superficie laevi, primum ab albida ad pallide flavam deinde stramineam, brunneo-flava, brunneo-ferruginosa vel brunneo-cuprea detersione maculata, ocraceo-rubra in vetustate; rhizomorphis appressis, sparsis vel plurimis. Gleba gelatinosa firma, sectione deliquescens; lacunae irregulares, flavo-viridescentes deinde ocraceo-rubrae, celeriter brunneo-rubrae sectione fientes, venis albis cinctae irregulare reticulum formantibus; pseudocolumella absens; odor nullus vel acidulus, serius acidus. Peridium cum duobus stratis: peridiopellis 10-50 µm spissa, hyphis appressis cum pigmento intraparietali, dermatocystidia sparsa vel abundantibus, 35-65 × 4-12 µm cum aut sine mucilagine emittentibus; subpellis 80-330 µm spissa, globulosa-prismatica textura cum cellulis 20-65 × 8-45 µm flavo spissoque pariete hyphis filamentosis sparsis. Gleba gelifica lacunis gelificis 250-600 µm latis, frequentibus 35-55 × 8-45 µm intercellulis. Basidiosporae 4.4-5.6 × 2.2-2.8 µm, pallide luteae, cylindricae, interna facie a plana ad leviter concavam obliqua imagine, biguttulatae; apicali plaga 1-1.2 µm lata sine reliquiis sterigmatorum. Basidia clavata, sessilia, 9-35 × 4.5-6.2 µm. Fibulae praesentes sed parum aspectabiles. Socius Alno alnobetulae subsp. alnobetulae alpino gradu, in Alpibus occidentalibus, et A. alnobetulae subsp. suaveolens in Corsica, frequens.

Holotypus: Savoie, Bourg-Saint-Maurice, 26.VIII.2007, coll. P.-A. Moreau n° 07082629; in herbario LIP conservatur; isotypus in herbario ZT.

Description of alpine material (PAM07082629): **Basidiomata** 0.8-2.5 × 0.8-1.5 cm, hypogeous to emergent, globose to subglobose, more rarely irregularly elongated to constricted, with usually a small but distinct basal spot from which some brownish rhizomorphs arise; surface smooth, at first pale yellowish to whitish, then straw-yellow, olivaceous, ochre-yellow, spotted of ferruginous-brown to copper-brown patches when bruished or with age, reddish ochre when dried or alterated; rhizomorphs scarse to abundant, adpressed on the surface, concolorous with peridium or darker especially the thickest ones around base. **Gleba** gelatinous-firm to soft when mature, when young whitish then gradually greenish yellow, olivaceous yellow, reddish-ochre to amber-brown with age, becoming pinkish-brown to reddish-brown after 20-60 seconds when cut, with intermixed white veins forming an irregular reticulum; pseudocolumella absent; on exsiccatum: amber-reddish, vitrified, with abundant, rather thick white veins (amber yellow on cut surface). **Smell** none to weakly sourish, acetic when old.

Peridium 2-layered. Peridiopellis 10-20 µm thick, amber-yellow, made of 1-2 discontinuous layers of cylindrical to clavate appressed hyphae, 4.5-13 µm wide, smooth, with thickened wall up to 0.8 (1) µm with pale yellow intraparietal pigment, with rather abundant cylindrical, attenuate to subcapitate dermatocystidia 35-65 × 4-12 µm, some covered by yellowish mucus or with thickened wall at apex, also forming occasional rhizomorphs made of cylindrical hyphae with thick yellowish wall up to 2-2.5 µm and covered by pale cristalloid incrustations. **Subpellis** 220-330 µm thick, yellow, of *textura globosa* to *textura prismatica*

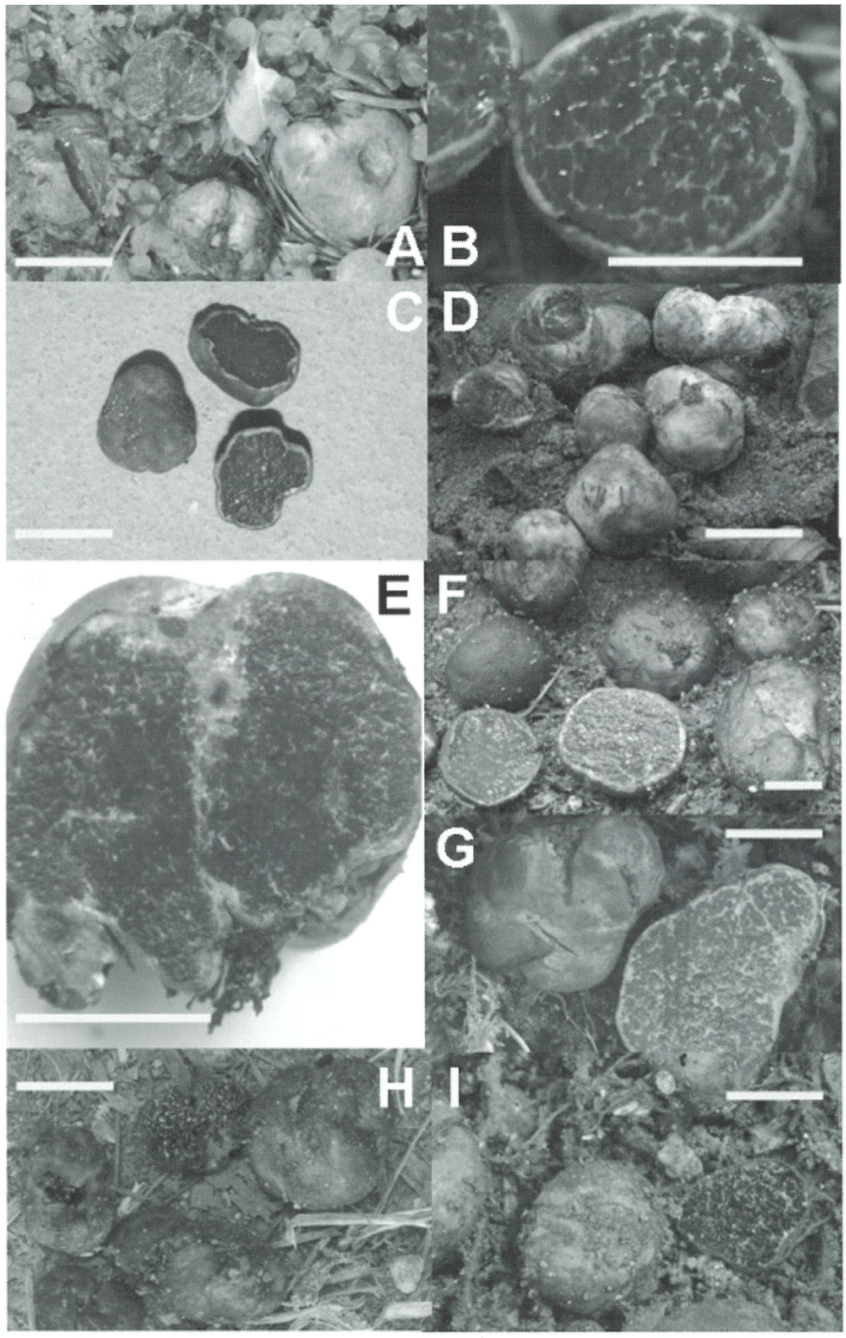

Fig. 1. Basidiomata. **A.** *Alpova alpestris*, coll. 07082629 (holotype, Alps); **B.** *Alpova alpestris*, coll. 08090302 (Corsica); **C.** *Alpova corsicus*, coll. 04101021 (holotype); **D.** idem, coll. FR27; **E.** idem, coll. 04101117; **F.** *Alpova* sp. (cf. *A. cinnamomeus*), coll. 09082702; **G.** *Melanogaster luteus,* coll. 09082801; **H.** *Melanogaster rivularis*, coll. 08090514 (holotype). – Scale bars = 10 mm. Photos P.-A. Moreau (A-D, F-H), F. Richard (E).

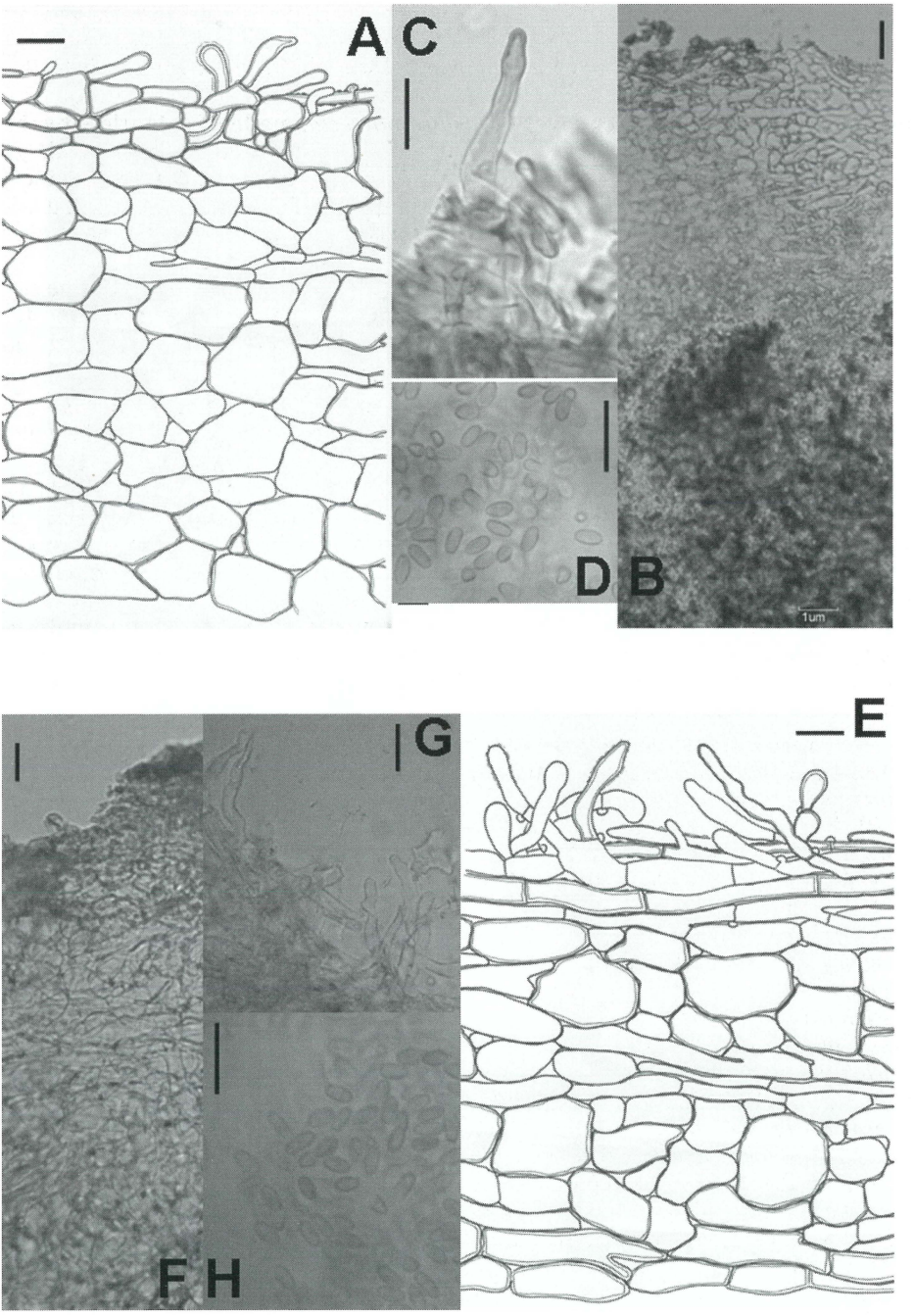

Fig. 2. **A-D.** *Alpova alpestris*, coll. 07082629 (holotype, Alps). A, B. peridium, radial cut; C. dermatocystidium on peridiopellis; D. spores. **E-H.** *Alpova alpestris*, coll. 08090302 (Corsica). E, F. peridium, radial cut; G. dermatocystidia on peridiopellis; H. spores. – Scale bars = 10 µm. Photos P.-A. Moreau.

structure (pseudoparenchymatous), made of short, rounded, ellipsoidal, pyriform or puzzle-like hyphae, 20-65 × 8-45 µm, with thick (1.8-2.5 µm) yellow wall, not gelified, without lacunes, occasionally mixed with cylindrical hyphae up to 90 × 8-25 µm, especially towards gleba; wall always smooth. Gleba separated from subpellis by a 30-40 µm-thick layer of *textura porrecta*, made of colourless hyphae 4-9 µm wide, made of slender hyphae mixed with wide, thin-walled and +/– anastomosing hyphae, all with slightly thickened yellow wall. **Gleba** made of gelatinized lacunae 250-600 µm wide, often ellipsoid 150-350 µm wide, filled with slender hyphae 2-3.5 µm wide, thin- to slightly thick walled, smooth, separated by a pale brownish context made of long, fewly septate cylindrical hyphae with thickened wall, 4-12 µm wide, wall 0.5-0.8 µm thick; buffer-cells[3] large, spherical-pedunculate, not unfrequent on mature specimens around locules, hyaline, often collapsed, 35-55 × 25-38 µm, with rather short peduncule up to 10-15 µm long directly issued from hyphae of veins oryidium.

Basidiospores (4.1) 4.4-*4.99*-5.6 (5.9) × 2.2-*2.46*-2.8 µm, Q = 1.82-*2.04*-2.27, V = 11.1-*16.06*-22.3 µm^3 [1 macrospore observed, 8 × 3.5 µm], pale yellow in KOH cylindrical, not strangulate in front view, most spores convex to obtuse on outer side towards base, inner side flat to slightly depressed (phaseoliform tendency) on side view, with a narrow and hardly distinct apical plage 1-1.2 µm wide, mostly biguttulate when mature, without rests of sterigmata; wall smooth, yellowish, slightly thickened (0.2-0.3 µm thick). **Basidia** 9-18 × 4.5-5.2 µm, clavate, sessile or with short base, not guttulate, 8-spores, thin-walled, with thin, hardly observable sterigmata, often with brownish content before maturity. **Subhymenium** ramose made of short catenulate articles, thin-walled, 2-3 µm wide, hyaline. **Clamps** not conspicuous but likely present at all septa. No part of the basidioma dextrinoid or amyloid.

Specimens examined. ROMANIA, Retezats Mts, Southern Carpathians, Retezatul Mic, Piule-Iorgovanu massif, slopes of Vârful Piule, tourist path under *Alnus viridis*, 1675 m, A. & M. *Ronikier*, 23 Aug. 2009, P.-A. Moreau 09082300 (LIP). France, Corse du Sud, Bastelica, val d'Ese, 1650 m, in raw peat under a living patch of *Sphagnum*, under *Alnus alnobetula* subsp. *suaveolens*, J. *Rochet* & P.-A. *Moreau*, 3 Sept. 2008, P.-A. Moreau 08090302; same locality, in a sandy river bank under *Alnus alnobetula* subsp. *suaveolens*, with *Alnicola submelinoides*, P.-A. *Moreau*, 10 Sept. 2005, P.-A. Moreau 05091024 (LIP); Haute-Corse, Vizzavona, Monte d'Oro, 1600 m, mixed peaty subalpine forest with *Fagus sylvatica* and *Pinus laricio*, under a single *Alnus alnobetula* subsp. *suaveolens*, F. *Richard*, 11 Oct. 2004, P.-A. Moreau 04101113 (LIP); same locality, under *Alnus alnobetula* subsp. *suaveolens* in a mesotrophic wet slope, 9 Sept. 2005, F. *Richard*, P.-A. Moreau 05102601 (LIP); Casamacchioli, lac de Nino, above the lake, wet thickets of *Alnus alnobetula* subsp. *suaveolens*, 1750 m, P.-A. *Moreau*, 5 Sept. 2007, P.-A. Moreau 07090501 (LIP); same locality, below the lake, 1750 m, under *Alnus alnobetula* subsp. *suaveolens* along a stream, P.-A. *Moreau*, 5 Sept. 2007, P.-A. Moreau 07090506 (LIP). Savoie: Bourg-Saint-Maurice, Arc 1800, 1650 m, in wet mineral-rich humus under *Alnus alnobetula* subsp. *alnobetula* and *Picea abies*, P.-A. *Moreau*, 26 Aug. 2007, holotypus herb. P.-A. Moreau 07082629 (LIP); same locality, P.-A. *Moreau*, 28 Aug. 2009, P.-A. Moreau 09082802 (LIP); same locality, 1950 m, along the golf course under *Alnus alnobetula* subsp. *alnobetula* on the humus-rich edge of a watercourse, with *Lactarius alpinus*, P.-A. *Moreau*,

3. Buffer cells: this term is taken from Zeller (1939) for the large rounded cells also observed by Nouhra *et al.* (2005), at the origin of the gelatinized lacunae (see Fig. 3H).

21 Aug. 2008, P.-A. Moreau 08082105 (LIP); Landry, loc. Barmont, 1580 m, under *Alnus alnobetula* subsp. *alnobetula* and *Picea abies* along a mineral-rich watercourse, with *Alnicola badia*, *Cortinarius bibulus*, *Lactarius alpinus*, *Russula alnetorum* etc., P.-A. *Moreau*, 26 Aug. 2007, P.-A. Moreau 07082632 (LIP); Les Saisies, Arrêté de Biotope de la tourbière des Saisies, 1560 m, on a wet road side under *Alnus alnobetula* subsp. *alnobetula*, P.-A. *Moreau*, 16 Aug. 1995, P.-A. Moreau 95081604 (LIP); Crest-Voland, Arrêté de Biotope de la tourbière des Saisies, along the Nant Bruyant stream, 1560 m, acidic peaty side under *Alnus alnobetula* subsp. *alnobetula*, P.-A. *Moreau*, 22 Aug. 2009, P.-A. Moreau 09082201, 09082202 (LIP); same locality, 1600 m, J. *Rochet*, 27 Aug. 2009, P.-A. Moreau 09082720 (LIP).

Ecology and distribution: so far only known from subalpine zone of the Alps, Carpathians and Corsica, under *Alnus alnobetula*, isolated in *Picea* forests as well as in pure stands, where it is widespread and locally abundant on all types of soils in wet places, usually along streams or watercourses amongst liverworts (*Pellia* spp.), between 1500 and 2100 m. In Corsica it is only reported under *Alnus alnobetula* subsp. *suaveolens* at subalpine level, between 1700 and 1900 m, in wet acidic soil, especially on peat and raw humus, where it is widespread but sparse, often single, and usually not or hardly emergent, easily overlooked in the field.

Observations: all analyzed samples from *Alnus alnobetula* from the Alps (but one, see *Alpova* sp. aff. *cinnamomeus*, below), coming from acidic peatlands, mesotrophic wetlands and basic spruce forests, seem to represent a single species, with a weak intraspecific genetic variability attributed here to populational variations. Corsican samples diverge from the former by minor micro-morphological characters, such as heterogenity of peridial structure (with frequent cylindrical hyphae and only locally with dominant globose elements), dermatocystidia plentiful, and long pedunculate basidia (9-18 × 4.5-5.2 µm for the Alps, 18-35 × 5-6.5 µm for Corsica). The collection studied from the Romanian Carpathians (09082300, LIP) differs from collections from Western Alps by a poorly developed peridium with voluminous cystidia, some of them clavate with brownish content and short clavate-capitate basidia (16-20 × 5.5-6.5 µm). More data from Central Alps, Carpathians and Balkans are required to precise its whole variability, geographic dispersion, and correlations with ecology or biogeography. Samples from the three sampled area (Alps, Carpathians and Corsica) show some genetic divergences (data not shown) but so far weakly supported, and do not currently justify a subspecific distinction.

In Corsica *A. corsicus* may be found in the same places as *A. alpestris*, and can be distinguished in the field by the whitish-tomentose peridium (on young specimens), gleba with regular globose lacunae, and *sub micr.* by absence of dermatocystidia and narrower spores (1.8-2.1 µm in width, vs. 2.1-2.4 µm for *A. alpestris*).

I.2. *Alpova corsicus* P.-A. Moreau and F. Richard, *sp. nov.*

Fig. 3D-F, 3A-C, 5K-M.

Mycobank MB 518392

Etymology: from Corsica island, where it is widespread and apparently endemic.

Basidiomata 1.2-4 cm in longitudine, 0.8-2.2 in diametro, a globosis ad irregulariter extensa, constricta vel lobata, basi distincta; superficie albida sericea deinde flavo-cremea, brunneo-ferruginea vel brunneo-cuprea detersione, brunnea obscura in vetustate; rhizomorphis sparsis. Gleba gelatinosa, sectione deliquescens; lacunae a sphaericis ad ellipticas, ab ocraceo-rubellis ad flavo-brunneas, brunneo-olivaceofuscae fientes post unam vel duas sexa-

gesimas partes horae, albis venis cinctae; pseudocolumella praesens vel flavae sterilique basali zonae limitata; laevis odor fungoideus, maturitate moschatus fiens. Peridium cum duobus stratis: peridiopellis 15-35 µm spissa, hyphis appressis parietali incrustantique pigmento, brevibus finibus sine distinctis dermatocystidiis; subpellis 150-200 µm spissa, globulosa-prismatica textura cellulis 20-45 × 5-30 µm, flavo spissoque pariete, hyphis filamentosis frequentibus. Gleba gelifica lacunis gelificis 150-600 µm latis, intercellulis 22-55 × 8-45 µm sparsis. Basidiosporae 4.5-5.6 × 1.9-2.5 µm, pallide flavae, anguste cylindricae saepe constrictae fronte visae, facie interna a plana ad concavam obliqua imagine, biguttulatae; apicali plaga 0.8-1 µm angusta parumque aspectabili, sine reliquiis sterigmatorum. Basidia clavata basi extensa, 12-20 × 3.5-4.5 µm. Fibulae praesentes sed parum aspectabiles. Socius speciebus variis Alni, in Corsica.

Holotypus: Alta Corsica, Ponte Leccia, sub Alno glutinosa, 10.X.2004, coll. P.-A. Moreau n° 04101021, in herbario LIP conservatur; isotypus in herbario ZT.

Description (PAM04101021): **Basidiomata** hypogeous to emergent, 1.2-4 × 0.8-2.2 cm, globose to irregularly elongated, often constricted or bilobate, with a distinct basal spot surrounded by small scarse rhizomorphs; surface at first whitish-silky the gradually cream yellow, then ferruginous-brown to copper-brown when bruished, dark brown when dried or alterated; rhizomorphs scarse, appressed on the surface, concolorous with peridium and quickly reddening when bruished. **Gleba** gelatinous-firm to soft when mature, when cut reddish-ochre, amber-brown, becoming dark olivaceous-brown after 1-2 minutes, with densely intermixed white veins; pseudocolumella variable, sometimes well-developed, only visible as a diffuse orange-yellow zone of 3-5 mm above the basal point; on exsiccatum: amber-reddish, vitrified, with abundant, rather thick white to pale yellow veins. **Smell** slightly fungoid when fresh, becoming musky-nauseous when very mature or during drying process.

Peridium 2-layered. Peridiopellis 15-35 µm thick, reddish-brown, made of 2-3 layers of coloured cylindrical hyphae, 5-9 µm wide, slightly incrusted to rough, with amber-yellow intraparietal pigment, with thickened wall up to 1.2 µm, also forming occasional rhizomorphs; occasionally emergent clusters of short hairs, up to 22-40 µm, 20-30 µm thick, multiseptate with terminal element shortly clavate and +/– curved, 6-9 µm wide, with thickened yellowish wall up to 0.5 µm, not incrusted. **Subpellis** 150-220 µm thick, pale yellow to almost colourless towards gleba, of heteromerous *textura globulosa* to *textura prismatica* structure (pseudoparenchymatous), made of short, rounded to ellipsoidal or shortly cylindrical hyphae, 20-45 × 5-30 µm, with rather thick (0.5-1.5 µm) pale yellow wall, amber-yellow to orange with thickened wall (up to 2.5 µm) towards surface, nor gelified, without lacunes, mixed with long hyphae, isolate or fasciculate by 3-4, of concentric orientation, hyaline, cylindrical 11-14 µm wide, especially towards gleba; wall always smooth. **Gleba** made of gelatinized lacunae 150-600 µm wide, globose to ellipsoid, filled with slender hyphae 2-3.5 µm wide, thin- to slightly thick walled, often rough, with a very thick gelatinous covering (multiguttulate aspect when revived in KOH + congo red), appearing like sinuose and very thick coscinoid hyphae, separated by a colourless context made of wide, strongly thick-walled hyphae 4.5-12 µm wide, wall 0.5-2.5 (3) µm thick, smooth, intermixed with sinuose thin-walled (coscinoid?) cylindrical hyphae 5.5-9 µm wide, with dense microguttulate content; buffer cells scattered around lacunae and directly attached to glebal veins, large, spherical-pedunculate, 22-55 µm wide, with peduncule up to 40 µm long. **Basidiospores** 4.5-*5.01*-5.6 × 1.9-*2.15*-2.5 µm, Q = 2.00-*2.35*-2.76, V = 8.8-*12.36*-17.5 µm^3 (but see the long-spored collection n° 07090813, Tab. 2) pale yellow in KOH, cylindrical, somewhate strangulate in front view on the most elongate spore, cylindrical to somewhat depressed on one face (phaseoliform tendency) on side view, with a rather narrow and inconspicuous apical plage

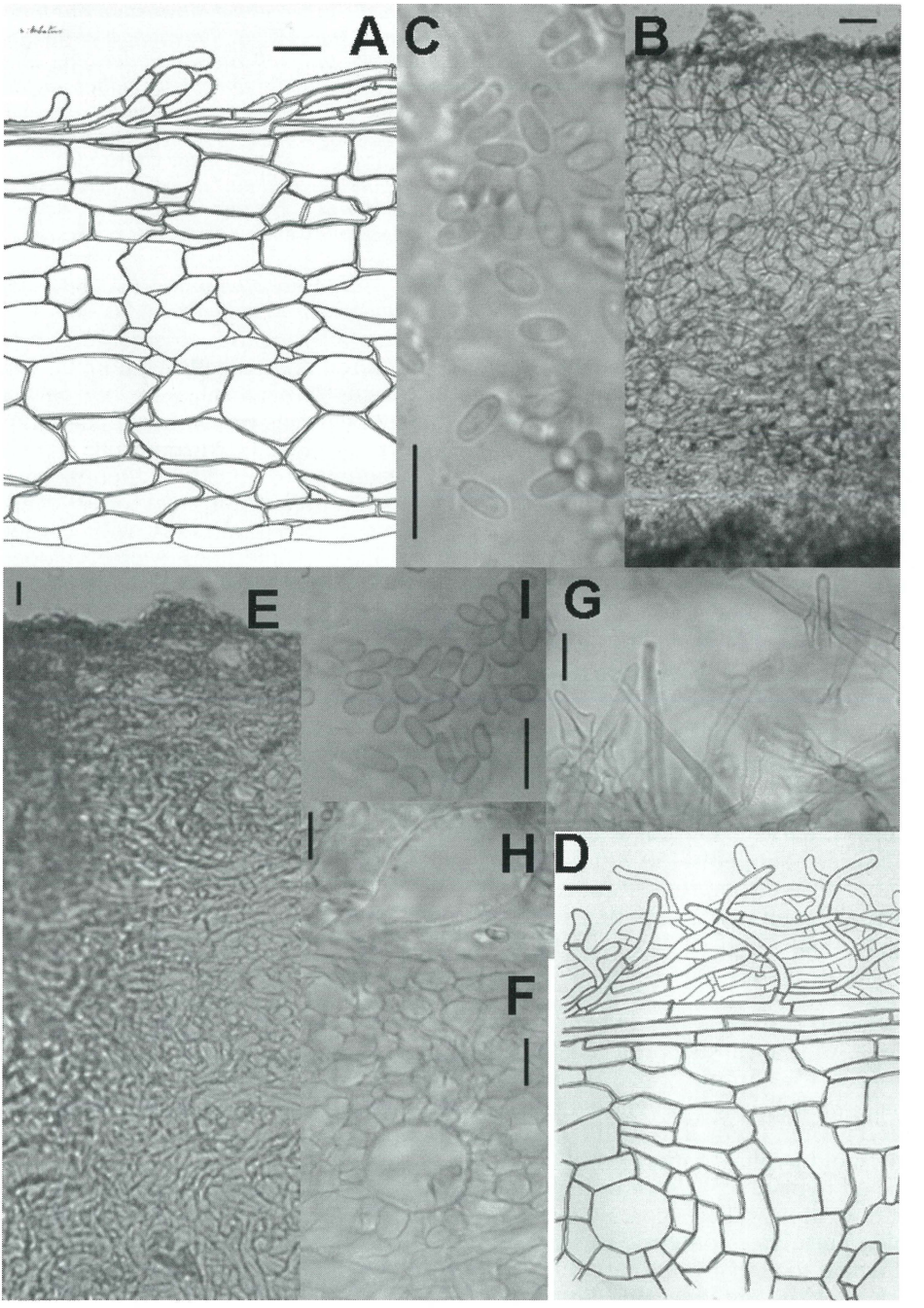

Fig. 3. **A-C.** *Alpova corsicus*, coll. 04101021 (holotype). A, B: peridium, radial cut; C: spores. **D-H.** *Alpova* sp. (cf. *A. cinnamomeus*), coll. 09082702. D, E: peridium, radial cut; F: detail of subpellis showing folliculoid structure (see derscription); G: detail of peridiopellis; H: buffer cell in gleba; I: spores. – Scale bars = 10 µm. Photos P.-A. Moreau.

0.8-1 (1.2) µm wide, mostly biguttulate when mature, without remnants of sterigmata; wall smooth, yellowish, slightly thickened (0.2-0.3 µm thick). **Basidia** 12-20 × 3.5-4.5 µm, clavate with elongate base, sparsely guttulate, early collapsed, mostly (8-9) 10 spores (when observable), thin-walled, with thin, hardly observable sterigmata. **Subhymenium** filamentous, with thin-walled hyphae 2-2.5 µm wide, with somewhat guttulate-refringent content. **Clamps** hardly distinct, but likely present at all septa. No part of the basidioma dextrinoid or amyloid.

Specimens examined. FRANCE, Haute-Corse, Ponte Leccia, Moltifao, tourbière de Valdo, 250 m, a river bank under *Alnus glutinosa*, in sand mixed with acidic peaty humus, with *Alnicola escharoides*, *Lactarius omphaliformis* and *Russula pumila*, P.-A. *Moreau* & F. *Richard*, 10 Oct. 2004, holotypus herb. P.-A. Moreau 04101021 (LIP); same locality, F. *Richard*, 13 Oct. 2008, FR27, FR28, FR29 (LIP); Rutali, Caratucucia, Forêt de Stella, 1030 m, in a pure *Alnus cordata* stand, with *Alnicola salabertii* and *Inocybe calospora*, F. *Richard*, 26 Oct. 2005, P.-A. Moreau 05102601 (LIP); Omita, Monte Estremo, Fango valley, 560 m, along a permanent watercourse under *Alnus cordata*, F. *Richard*, 12 Oct. 2008, FR1 (LIP); Verghello, along a stream on sandy alluvions under *Alnus glutinosa* and *A. cordata*, P.-A. *Moreau* & F. *Richard*, 11 Oct. 2004, P.-A. Moreau 04101117 (LIP); Corse du Sud, Bastelica, val d'Ese, 1650 m, in a sandy river bank under *Alnus alnobetula* subsp. *suaveolens*, P.-A. *Moreau*, 8 sept. 2007, P.-A. Moreau 07090813 (LIP) (long-spored form).

Ecology and distribution: so far only known from Corsica, where it is found fruiting abundantly in riparian and peaty forests under *Alnus glutinosa*, but also under *A. cordata* at supramediterran level, and more occasionally under *A. alnobetula* subsp. *suaveolens* (one single collection with longer spores: n° 07090813) where it may be confused with *Alpova alpestris*. This species shows a wide host-range, but was specifically associated with each *Alnus* species in environmental contexts corresponding to their preferential altitudinal belt (Gamisans 1993).

Observations: this species shows a wide distribution in Corsica, where it fruits under all species of *Alnus*, apparently mainly in acidic and mineral substrates. It seems to be well-characterized by narrower spores than other European species, but a collection found under *Alnus alnobetula* subsp. *suaveolens*, proved to belong to this species by molecular analysis (data not shown), showed much longer and wider spores; a relation with elevation is possible (e.g. reduction of number of sterigmata, hardly observable on *exsiccata*).

Unless conspicuous basidiomata (often large and completely emergent) are easily observed in Corsica, this species is not reported from continental *Alnus* forests and can be considered as a Corsican endemics at this stage of our knowledge; investigations in Sardinia and other Mediterranean stands of *Alnus glutinosa* or *A. cordata* are required.

I.3. *Alpova* sp. (cf. *A. cinnamomeus* C.W. Dodge). Fig. 1G, 3E-I, 5N-P.

Description (PAM09082702): **Basidiomata** semi-hypogeous, subglobose, 3.5 × 2 cm, with a distinct basal spot surrounded by brownish rhizomorphs; surface glabrous, ochre-yellow, then ferruginous-brown when bruished, dark reddish brown when dried; rhizomorphs abundant around the base but not elsewhere, concolorous with peridium, dark brown when dry. **Gleba** gelatinous-firm, not distinctly deliquescent, when cut brownish-grey, not distinctly turning, with densely intermixed white veins delimiting small, subglobose then confluent lacunae; pseudocolumella not distinct but some veins of radial orientation

reaching peridium, with an orange-yellow zone with small unmature lacunae above the basal point; on exsiccatum: blackish, not apparently vitrified, with a dense net of rather thick, pale yellow veins. Smell slightly fungoid with acetic component.

Peridium 2-layered. Peridiopellis 30-40 µm thick, reddish-brown, made of 4-7 layers of coloured cylindrical hyphae, 3.5-8 µm wide, smooth to slightly incrusted, with brownish intraparietal pigment, wall thickened up to 1.2 µm, also forming occasional rhizomorphs; around base (and probably elsewhere when very young) forming a thick tomentum up to 120 µm thick, early collapsed and adpressed but also visible as intermixed hyphae, similar to previous, multiseptate with terminal elements numerous, cylindrical, clavate to attenuate, 6-9 µm wide, not incrusted. **Subpellis** 180-250 µm thick, colourless to pale yellow, of *textura prismatica* structure (pseudoparenchymatous), made of short, polygonal-isodiametric to cylindrical hyphae, 15-70 (110) × 5-25 µm, with thin to moderately thick (0.3-1.5 µm) pale yellow wall, nor gelified, mixed with isolate or fasciculate cylindrical to fusiform hyphae of tangential orientation, 11-22 µm wide, and with locally numerous lacunae forming follicule-like structures at first, 15-25 µm diam.; wall always smooth. **Gleba** made of gelatinized lacunae 150-550 µm wide, ellipsoid to subglobose, filled with hardly distinct slender hyphae 2.5-4 µm wide, thin- to strongly thick walled, often rough, strongly gelified, multiguttulate in KOH, separated by a colourless context of rather wide veins up to 100-250 µm diam., made of intermixed colourless to pale yellowish hyphae, 3-6.5 µm wide, mostly short and ramose especially in nodes, and occasionally with long cylindrical thick-walled hyphae 9-18 µm wide, wall 0.5-1.5 µm thick, smooth; buffer cells abundant especially along subpellis but frequent around lacunae and directly issued from wide hyphae 3-4.5 µm wide along the veins, large, ovoid- to spherical-pedunculate, 18-45 µm wide, more or less pedunculate. **Basidiospores** (3.8) 4.4-*4.93*-5.4 (6.0) × 2.1-*2.33*-2.5 (3.11) µm, Q = 1.83 -*2.13*-2.49, V = 10.9-*14.21*-17.4 µm^3, pale grey-ochre in KOH, cylindrical, occasionally slightly strangulate in front view on the most elongate spore, cylindrical to often slightly depressed on one face in side view (phaseoliform tendency, rarely boomerang-shaped), with a narrow and inconspicuous apical plage less than 0.8 µm wide, mostly biguttulate of with 2 false septa when mature, without remnants of sterigmata; wall smooth, yellowish, slightly thickened (0.2-0.3 µm thick), sometimes thickened towards base. **Basidia** 8-14 × 3.5-5 µm, clavate with short to slightly elongate base, minutely guttulate before maturity, early collapsed, mostly (6) 7-8 spores (when observable), thin-walled, with thin, hardly observable sterigmata. Subhymenium filamentous, with thin-walled hyphae 2-2.5 µm wide. **Clamps** very small and often hardly distinct, but likely present at all septa. No part of the basidioma dextrinoid or amyloid.

Specimens examined. France, Savoie, Beaufort, cormet d'Arèches, a single basidiome under *Alnus alnobetula* subsp. *alnobetula*, semi-hypogeous in a wet mineral slope on calcic schist, with *Lactarius lepidotus*, *Cortinarius bibulus*, *Alnicola badia* and *A. inculta*, P.-A. *Moreau*, 27 Aug. 2009, P.-A. Moreau 09082702 (LIP).

Observations: This unexpected collection of a single basidiome was found in a pure *Alnus alnobetulae* slope on very mineral substrate (alterated calcschists and gypsus), rich in ectomycorrhizal basidiomata (dominated by *Lactarius lepidotus*) where no *Alpova* had been reported before despite of careful and repeated investigations. Morphologically this basidiome was much larger than usual *Alpova alpestris*, the peridium was almost devoid of rhizomorphs except around a rather well-differentiated base, and the gleba did not turn reddish when cut. Abundance of buffer cells and heterogenous structure of subpellis, and very

small unconspicuous basidia make it close to *A. corsicus* and especially also to *A. austroalnicola*. The apparently biseptate spores is an unusual character but the systematic value of this observation must be confirmed on more collections.

This species seems to be even closer to a collection from Sweden (Genbank accession number: DQ989497, see Fig. 7), found under *Alnus incana* (M. Berbee, pers. com.). The existence of another, yet unnamed species associated with *Alnus incana* in Europe, unknown to us, is therefore probable and requires further investigations.

II. Genus *Melanogaster* Corda, Deutschlands Flora, Abt. III. Die Pilze Deutschlands 3 (11): 1. 1831.

Remarks: Knapp (1954a: 117) proposed a subdivision of the genus *Melanogaster* into three "groups", respectively called *Ambiguus*-group (defined by spore length > 10 µm), *Variegatus*-group (spore length between 7 and 10 µm), and *Microsporus*-group (spore length < 7 µm). Svrček (1958) arises this division at rank of Sections, without validation by latin diagnosis, followed by Pegler *et al.* (1983). Our analysis (Fig. 7), following those of Halász (2008), tends to confirm the pertinence of Knapp's proposal for European species on traditional microscopic basis. As far as we know these infrageneric names have never been validated, therefore we propose only provisionally the following sectioning for European species of *Melanogaster*, without presuming the position of extra-European species in this systematics:

– Section *Melanogaster* (= sect. *Ambigui* Svrček 1958: 532, inval.): including the type species: *M. tuberiformis* Corda, and also *M. ambiguus* (Vitt.) Tul., *M. intermedius* (Berk.) Zeller & C.W. Dodge, and *M. macrosporus* Velen., all with long spores (more than 10 µm length in average).

– Section *Variegati* Svrček 1958: 540 (inval.) (= section *Microspori* Svrček 1958, inval): including at least *M. broomeanus* Berk., *M. odoratissimus* Soehner & Knapp, *M. microsporus* Velen., *M. variegatus* (Vitt.) Tul., and *M. vittadinii* Soehner & Knapp, with spores of 6-10 µm average length, not associated with *Alnus* (see Halász, 2008, for comparisons and possible synonymies).

– Section *Rivulares* P.-A. Moreau, *ad int.*: including small-spored species of 5-6 µm average length, associated with *Alnus*: *M. luteus* Zeller and *M. rivularis sp. nov.*

II.1. *Melanogaster luteus* Zeller, *Mycologia* 31: 9. 1939. Fig. 1I, 4E-H, 5C-D.

= *Alpova luteus* (Zeller) Trappe, *Beih. Nova Hedwigia* 51: 291. 1975.

= *Melanogaster microsporus* Mattir., *Beitr. Kryptogamenfl. Schweiz* 8: 39. 1935 (illegit., non *M. microsporus* Velen. 1922).

= *Alpova diplophloeus* f. *europaeus* Trappe, *Beih. Nova Hedwigia* 51: 289. 1975.

Excluded: *Melanogaster luteus* sensu Zeller (1939: 10) and *Alpova luteus* sensu Trappe (1975: 291), which are '*Alpova*' *trappei* (Fogel 1977).

Illustrated references: Mattirolo (1935: 37-38, pl. II, as *M. microsporus*); Kers (1981, as *A. diplophloeus*); Montecchi and Lazzari (1989: 45-47, as *A. diplophloeus*); Montecchi and Sarasini (2000: 402-404, as *A. diplophloeus*).

Description (PAM09082801): **Basidiomata** 0.7-1.2 × 0.4-1,0 mm, hypogeous, globose, subglobose to ellipsoidal somewhat flattened, without distinct basal zone. Surface bright gold-yellow, wholly-felty sub lente, on dark reddish brown ground early visible when bruished or eroded; no rhizomorphs visible. **Gleba** gelatinous, early deliquescent when mature, initially black, not changing

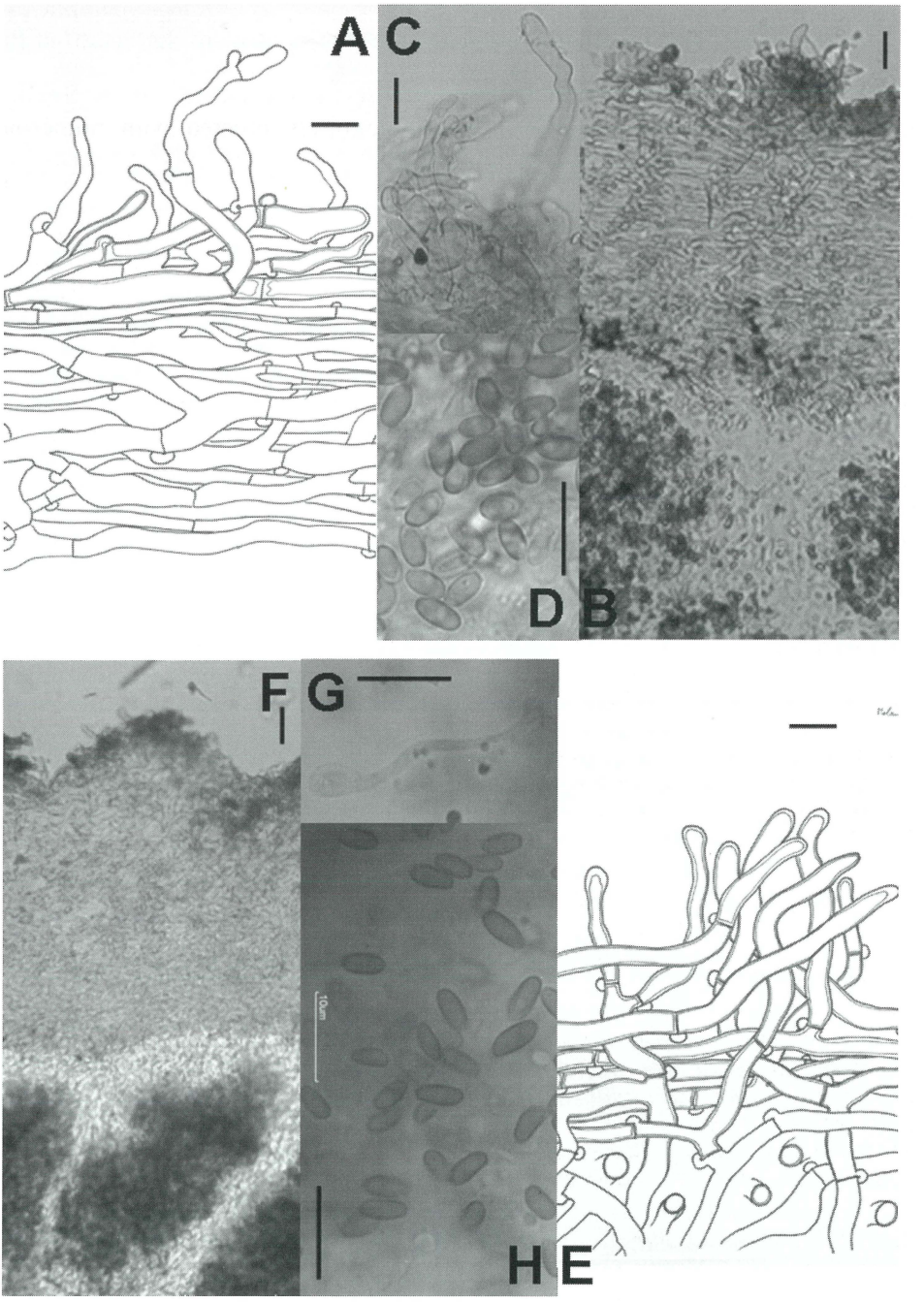

Fig. 4. **A-D**: *Melanogaster rivularis*, coll. 08090514 (holotype). A, B: peridium, radial cut; C: detail of peridiopellis; D: spores. **E-H.** *Melanogaster luteus*, coll. 09082801. E, F: peridium, radial cut; G: young basidium; H: spores. – Scale bars = 10 µm. Photos P.-A. Moreau.

when cut, with thin white veins forming a confuse net; pseudocolumella not distinct; on exsiccatum: black, vitrified, veins becoming cream to yellowish. Smell weak on mature specimens, of tar.

Peridium 2-layered. Peridiopellis 100-180 (250) µm thick, made of intermixed coloured hyphae, 4.5-6.5 µm wide, confusely erected with numerous terminations, smooth to slightly granulose, with dark amber-brown to reddish-brown intracellular pigment, with thickened colourless wall up to 1 (1.5) µm thick; terminal articles with rounded, attenuate to clavate apex, sometimes strongly thickened at apex and covered by +/– distinct colourless mucoid secretions. **Subpellis** 180-250 µm thick, pale greyish brown, of *textura porrecta* (prosenchymatous structure), made of slender hyphae 3-7.5 µm wide, with some inflated hyphae up to 12 µm wide, with pale yellow wall 0.3-0.5 µm thick, multidirectional, more parallel around lacunae, forming small gelatinized lacunes 20-40 µm wide; wall smooth, no incrustation seen. An abundant red-brown pigment is extractible in KOH. **Gleba** made of gelatinized lacunae 120-600 µm wide, globose to irregularly ellipsoid in cut, separated by veins made of gelatinized, long cylindrical hyphae 4.5-6 µm wide, often with thin incrusting pigment, thin-walled to slightly thick-walled, often collapsed; buffer cells not seen. **Basidiospores** (4.7) 5.3-*6.01*-6.8 (7.6) × (2.2) 2.4-*2.75*-3.0 (3.5) µm, Q = 1.95-*2.20*-2.50, V = 16.8-24.17-31.7 µm^3, grey-brown in KOH, with a large apical base 1.5-1.8 (2) µm wide, asymetric in profile view, somewhat inflate to club-shaped, the largest more elongate with obtuse to attenuate apex and sometimes "S" shaped, cylindro-elliptical to cylindrical on face view, mostly biguttulate when mature, with very short remnants of sterigmata; wall smooth, < 0.2 µm thick. **Basidia** 18-24 (32) × 5-6.5 µm, clavate-capitate when young, with more or less elongate peduncule, with 2-6 guttules when mature, mostly 8-spores, thin-walled and collapsed when mature, with short cylindrical sterigmata. **Subhymenium** filamentous, with thin-walled hyphae 1.5-5.5 µm wide, coarsely gelified, smooth; thin-walled, often microguttulate. **Clamps** wide and conspicuous, present at all septa. No part of the basidioma dextrinoid or amyloid, but peridiopellis naturally dark red brown and subpellis staining straw yellow in Melzer's reagent.

Specimens examined. FRANCE, Savoie, Hauteluce, towards Belleville, hypogeous, between the roots of a young *Alnus incana* on dark mineral alluvions, along a stream, alt. 1180 m, M. *Gardes*, P.-A. *Moreau* & J. *Rochet*, 28 Aug. 2009, P.-A. Moreau 09082801 (LIP); MONTE NEGRO, Opasanica, river side under *Alnus incana* and *Fagus moesiaca*, leg. B. *Perič*, 20 July 2006 (LIP and herb. pers. B. Perič).

Ecology and distribution: originally found in a mixed *Castanea-Pinus* forest in Northern Italy (Piemonte), where the presence of *Alnus glutinosa* is probable; documented here on two collections of sporocarps and a supplementary mycorrhizal sampling, under *Alnus incana* on river banks (France and Montenegro). Rarely reported but possibly overlooked in suitable habitats. Probably also present in Sweden (Kers 1981), under *Alnus glutinosa*.

Observations: this species possibly has a strictly continental-mountainous distribution, since it has never been found with *Alnus glutinosa* in lowland oceanic or subcontinental areas despite of numerous mycorrhizal samplings and field observations. It can be compared morphologically with insufficiently documented extra-European species for which no DNA sequence is available yet: *Melanogaster ovoidisporus* Y. Wang from China (Wang *et al.* 1995) and *Melanogaster minysporus* Cázares *et al.* from *Quercus* forests in Mexico (Cázares *et al.* 2008), which affinities seem closer to *M. variegatus* due to ovoid spores and habitat.

II.2. ***Melanogaster rivularis*** P.-A. Moreau & F. Richard, *sp. nov.*
Fig. 1I, 4A-D, 5A-B.

Mycobank MB 518393

Etymology: "riparian", found on brook and river sides.

Basidiomata 0.6-2.5 cm in longitudine, 0.4-1.8 cm in diametro, globosa vel cum duobus vel tribus lobis, basi depressa paene distincta; superficie tenuiter furfuracea, a flavo-aurata ad flavo-brunneam, deinde a brunneo-cuprea ad brunneo-rubram detersione; rhizomorphis absentibus. Gleba gelatinosa firma; lacunae a sphaericis ad ellipticas, atra, sine coloris mutatione sectione, albis tenuibusque venis cinctae; pseudocolumella distincta specie parvae basalis sterilis flavo-aurantiacae zonae; odor nullus, exiliter moschatus dessicatione. Peridium cum duobus stratis: peridiopellis 20-35 µm spissa, hyphis mixtis intraparietali brunneo-flavo pigmento interdum incrustanti, fasces pilorum brevium formantibus; subpellis 70-100 µm spissa, textura porrecta tenuibus hyphis pallide flavis, levibus vel leviter incrustatis, cum lacunis gelificis glebam versus. Basidiosporae 5.1-6.2 × 2.7-3.1 µm, griseo-brunneae, a cylindrico-ellipticis ad cylindricas fronte visae, asymmetricae amygdaliformesque obliqua imagine, biguttulatae; plaga apicali 1.5-1.8 µm lata, obliqua, cum reliquiis brevium sterigmatorum. Basidia 18-26 ×5.5-6.5 µm breviter clavata, pariete aliquanto spisso. Fibulae evidentes. Socius variis speciebus Alni, in Corsica.

Holotypus: Alta-Corsica, Asco, sub Alno glutinosa, 5.IX.2008, coll. P.-A. Moreau n° 08090514, in herbario LIP conservatur; isotypus in herbario Z+ZT.

Description (PAM08090514): **Basidiomata** 0.6-2.5 × 0.4-1.8 cm, hypogeous to half-emergent, globose to subglobose, often 2-3-lobate, base slightly depressed with hardly distinct basal zone. Surface at first gold-yellow to amber-yellow, minutely furfuraceous, early eroded, on copper-brown to reddish-brown ground, becoming darker when bruished; no rhizomorph visible. **Gleba** gelatinous-firm, anthracite-black, without distinct change when cut, with thin white veins intermixed; pseudocolumella very short but distinct in most specimens, reduced to a diffuse orange-yellow zone of 3-5 mm above the basal point; on exsiccatum: black, vitrified with thin white cotton-like veins. **Smell** none when fresh, somewhat musky when drying, weak.

Peridium 2-layered. Peridiopellis 20-35 µm thick, made of 2-4 layers of +/– intermixed or contorted coloured hyphae, 3.5-8 (11) µm wide, smooth to sometimes slightly incrusted, with brown-yellow intraparietal pigment (possibly also intracellular), the thinnest on the surface, sometimes paler and with thickened wall up to 0.5 µm; occasionally emergent clusters of short hairs, up to 50-90 µm thick, multiseptate with terminal elements attenuate to cylindro-clavate, 5-11 µm wide, with thickened yellowish wall up to 0.3 (0.5) µm, sometimes incrusted. **Subpellis** 70-100 µm thick, colourless, of *textura porrecta* (prosenchymatous structure), made of slender hyphae 3.5-5 µm wide, with pale yellow wall 0.3-0.5 µm thick, parallel towards surface, more multidirectional with some gelatinized lacunes 10-15 µm wide towards gleba; wall smooth to slightly punctate, more incrusted in the transition to gleba. **Gleba** made of gelatinized lacunae 80-160 µm wide, globose to ellipsoid in cut, separated by 4-6 layers of long cylindrical hyphae 2.5-6 µm wide, colourless in KOH, thin-walled to slightly thick-walled, often collapsed on mature specimens; buffer cells not seen. **Basidiospores** (4.2) 5.1-*5.65*-6.2 × 2.7-*2.91*-3.1 µm, Q = 1.74-*1.95*-2.17, V = 20.5-*25.25*-31.0 µm^3, grey-brown in KOH, somewhat asymetric with a large +/– oblic apical base 1.5-1.8 µm wide, cylindro-elliptical to cylindrical, with an amygdaloid tendency in profile view, mostly biguttulate when mature, without remnants of sterigmata; wall smooth, < 0.1 µm thick. **Basidia** 18-26 × 5.5-6.5 µm, shortly clavate, always hyaline, mostly 6-8-spores, thick-walled in mid upper part when mature, with short rounded thick-walled sterigmata before spore formation. Subhymenium filamentous, with thin-walled hyphae 2-3.5 µm wide, coarsely gelified and hardly distinct. **Clamps** wide and conspicuous, present at all septa. No part of the basidioma dextrinoid or amyloid.

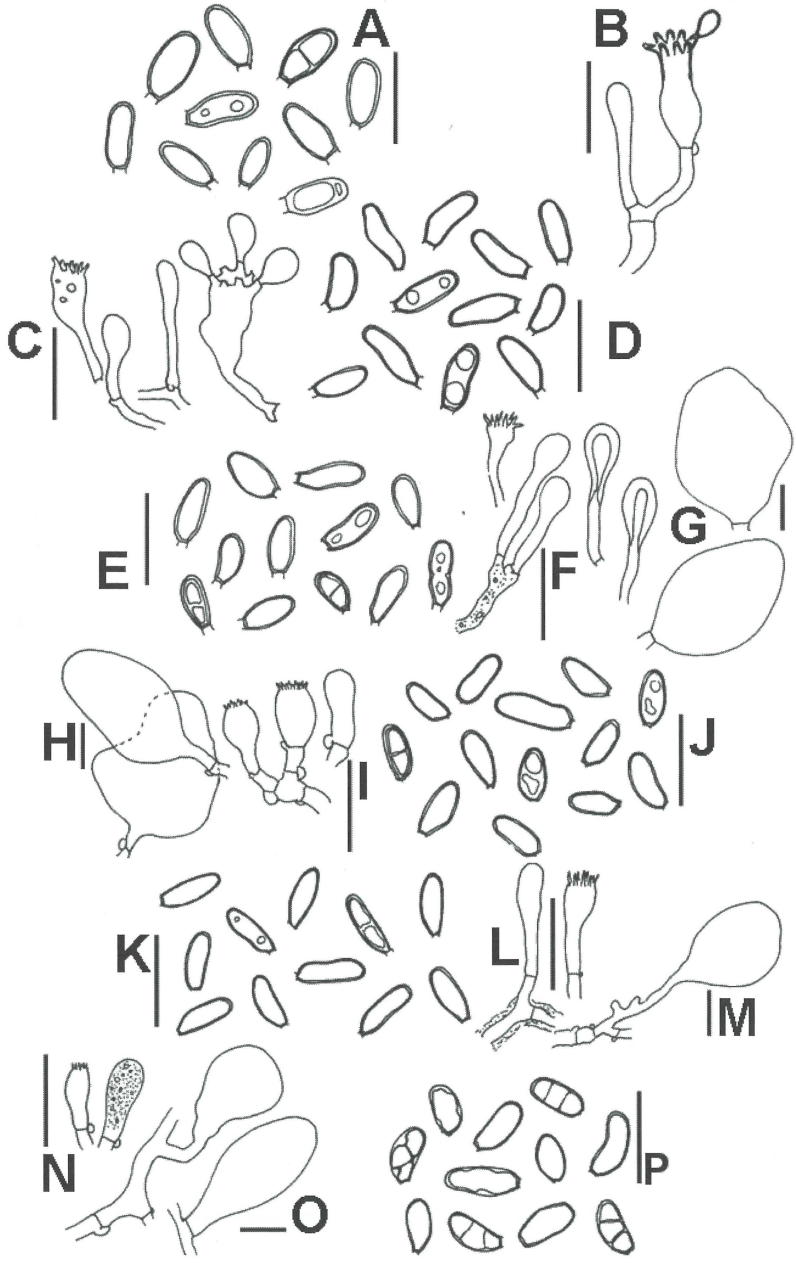

Fig. 5. **A-B.** *Melanogaster rivularis*, coll. 08090514 (holotype). A: spores; B: basidia. **C-D.** *Melanogaster luteus*, coll. 09082801. C: basidia; D: spores. **E-G.** *Alpova alpestris*, coll. 07082629 (holotype, Alps). E: spores; F: basidia and young sclerobasidia. G: buffer cells. **H-J.** *Alpova alpestris*, coll. 08090302 (Corsica). H: spores; I: basidia; J: buffer cells. **K-M.** *Alpova corsicus*, coll. 04101021 (holotype). K: spores; L: basidia and gelatinized subhymenial hypha; L: buffer cell. **N-P.** *Alpova* sp. (cf. *A. cinnamomeus*), coll. 09082702. N: spores; O: basidia; P: buffer cells. – Scale bars = 10 mm.

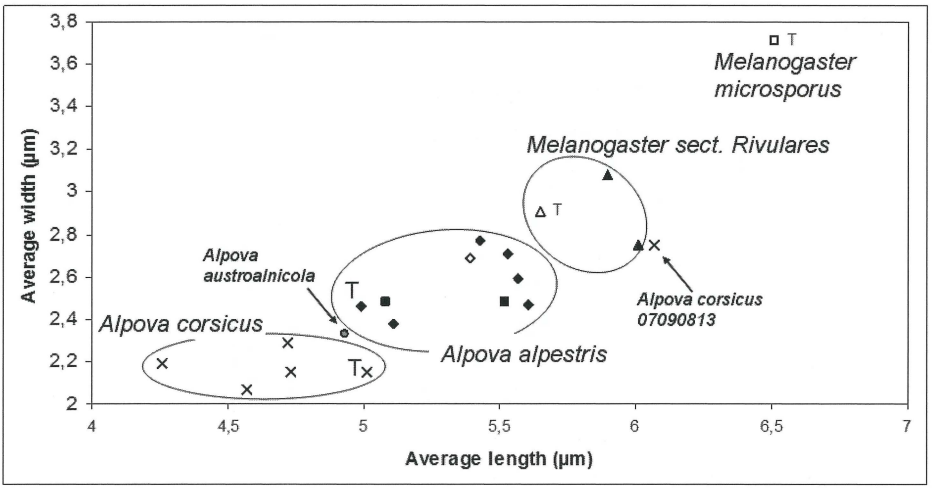

Fig. 6. Average spore dimensions of studied collections (see Table 2 for values). ◆: *Alpova alpestris* (Alps); ■: *A. alpestris* (Corsica); ◇: *A. alpestris* (Romania); ×: *A. corsicus*; ○: *A. austro-alnicola*; ▲: *Melanogaster luteus*; △: *M. rivularis*; □: *M. microsporus* Velen. T: holotype collections.

Specimens examined. FRANCE, Haute-Corse, Asco, under a young *Alnus glutinosa* not far from *Alnus cordata*, in a sand bank of a mountain stream, alt. 1650 m, P.-A. *Moreau* and J. *Rochet*, 5 Sept. 2008, holotype P.-A. Moreau 08090514 (LIP); Sant'Andrea-di-Boziu, under *Alnus cordata*, leg. S. *Biancardini*, L. *Hugot* and J. *Rochet*, 8 Oct. 2008, FR4 (LIP).

Ecology and distribution: only known from Corsica, where it is widespread and locally abundant under all species of *Alnus*, especially in stream beds amongst fine gravels and pebbles. The number of mycorrhizal samplings suggest this species to be locally dominant in root systems of alders, especially *Alnus cordata*. According to ecological data, this species preferentially associates with hosts that were at the upper limit of their altitudinal distribution in Corsica island (i.e. the mountain belt for both *A. glutinosa* and *A. cordata*; Gamisans 1993).

Observations: this Corsican species can be confused in the field with *Alpova* species (*A. alpestris* and *A. corsicus*) that can grow in the same places; it differs by a darker gleba without greenish-olivaceous tinges, furfuraceous surface, absence of staining when cut and lack of rhizomorphs. *Sub micr.* it is easily distinguished by a filamentous peridium structure without globose elements. It differs from *A. luteus*, a continental species, by much larger dimensions and dull peridial colours. The thick-walled basidia seem to be a distinctive feature for this species.

III. Excluded or doubtful taxa based on European material

III.1. *Alpova klikae* (Mattirolo) Trappe, *Beih. Nova Hedwigia* 51: 303. 1975.

≡ *Cremeogaster klikae* Mattirolo, *Atti Reale Accad. Sci. Torino* 69: 240. 1934.

Observations: the neotype designed by Trappe (1975: 304), identified by Mattirolo, has not been revised. Trappe (*op. cit.*) described this collection (preserved in TO) with a strong sporal variability, including large thick-walled

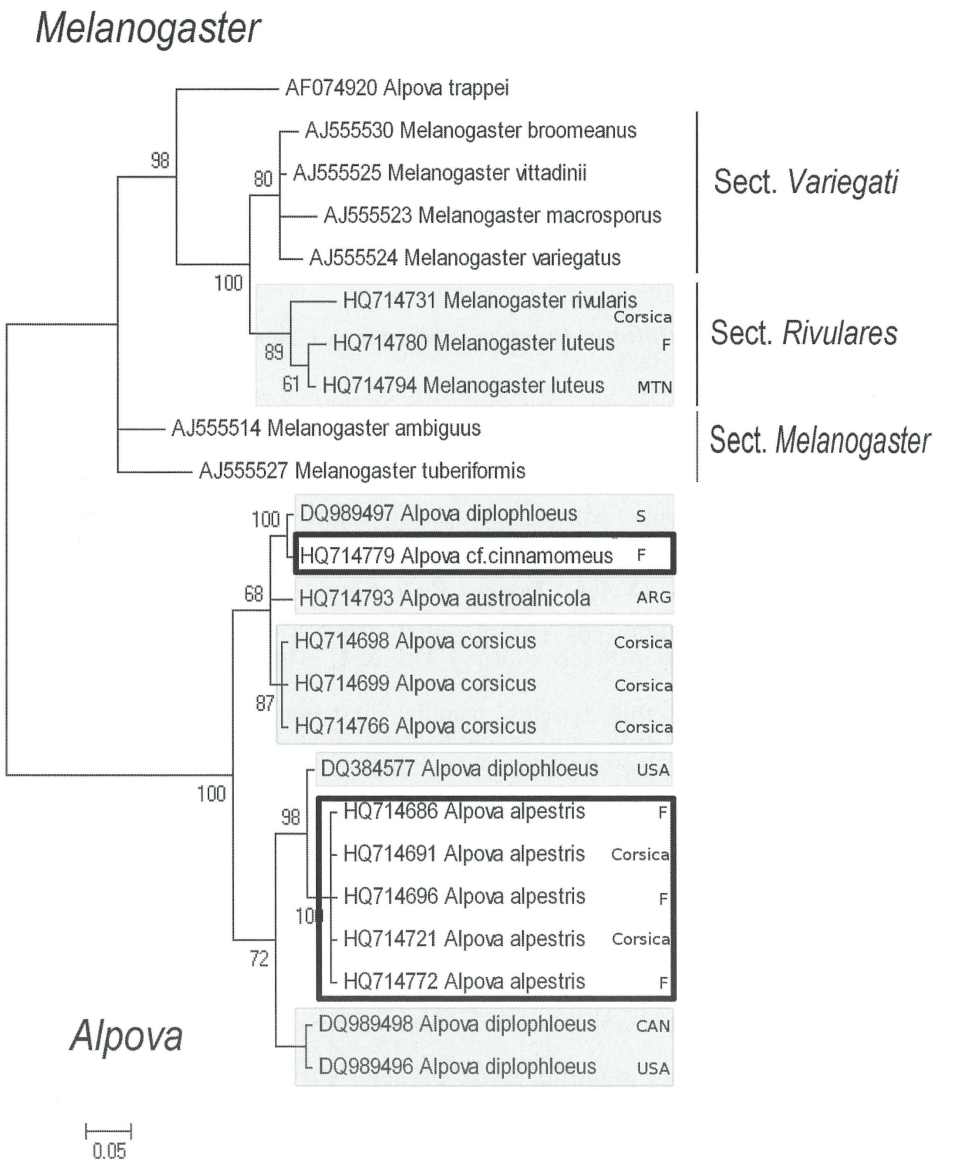

Fig. 7. Phylogenetic reconstruction of the *Alpova-Melanogaster* lineage. The 50% majority rule consensus tree inferred from Bayesian inference is presented. Numbers at nodes represent Bayesian posterior probabilities. In grey: species associated with *Alnus* subgen. *Alnus*. Circled: species associated with *Alnus* subgen. *Alnobetula*. Abbreviations: ARG: Argentina; CAN: Canada; F: France; MTN: Montenegro; USA: United States of America.

spores interpreted by him as chlamydospores. The hypothesis of a heterospory due to paucispored basidia of a more usual *Melanogaster* species (possibly *M. luteus*), cannot be rejected currently. The indications of locality ("Rodero, near Como") are curiously the same as those given for *Melanogaster microsporus* Mattirolo (= *M. luteus*).

The unique report of *A. klikae* by Schmid-Heckel (1987) and Gross (1980), found under *Alnus alnobetula* in Bavaria and identified on the basis of "chlamydospores", evokes an immature or substerile collection of *Alpova alpestris* (with whitish gleba and "sclerobasidia", but with typical pseudoparenchymatous peridium); one of our collections (see Fig. 2) also showed sclerobasidia but normally coloured spores.

III.2. ***Alpova pseudostipitatus*** **Calonge & Siquier ex Calonge & Siquier**, *Bol. Soc. micol. Madrid* 20: 301. 2000.

= *Alpova pseudostipitatus* Calonge & Siquier, *Bol. Soc. micol. Madrid* 18: 93. 1998 (invalid).

Observations: the original illustrated description (Calonge and Siquier 1998) clearly shows a basidiome with white hollow columella, and an olivaceous gleba not forming lacunae, which excludes this species from any genus currently recognized in *Boletales*. This species most likely belongs to the order *Phallales*.

III.3. ***Alpova rubescens*** **(Velen.) Trappe**, *Beih. Nova Hedwigia* 51: 294. 1975.

≡ *Octaviania rubescens* Vitt., *Monogr. Tuberacearum*: 18. 1831.

≡ *Melanogaster rubescens* (Vitt.) Tul. & C. Tul., *Hist. Monogr. Champ. Hypogés*: 96. 1851.

Observations: this species usually classified in *Melanogaster* sect. *Melanogaster* (Knapp 1954a; etc.) seems to have an ambiguous position between *Alpova* and *Melanogaster* (Vizzini et al. 2010). The pseudoparenchymatous peridium described by these authors is apparently typical for *Alpova* sensu stricto but the elongate spores (more than 10 µm long) with long appendages, the strong smell, the association with *Fagaceae*, and the apparent absence of buffer cells are general characters of *Melanogaster*. Vizzini (pers. com.) suggests that this species might represent a third lineage (genus) of gastroid *Paxillaceae*.

III.4. ***Alpova rubescens*** **var.** ***obscuratus*** **(Svrček) Trappe**, *Beih. Nova Hedwigia* 51: 294. 1975.

≡ *Melanogaster rubescens* var. *obscuratus* Svrček, Flora ČSR B1: 797. 1958.

Observations: a doubtful taxon (Trappe 1975; 294), possibly closer to typical *Melanogaster* species because of spore colour (Svrček 1958: 540, 796). A revision of the type material is required.

III.5. ***Melanogaster microsporus*** **Velen.**, *České Houby* 4-5: 809. 1822.

≡ *Alpova microsporus* (Velen.) Trappe, *Beih. Nova Hedwigia* 51: 302. 1975.

Observations: revision of type collection clearly shows that this species is distinct from all taxa described above. Plectenchymatous subpellis and spore dimensions (cf. Table 2) place it in the genus *Melanogaster* sensu stricto, and no indication given in the protolog (Velenovský 1922) evokes a possible relation with *Alnus* sp. Whether *M. microsporus* is a good species or a variant of *M. broomeanus* or *M. variegatus* (both hardly separated by microscopical and molecular data; see Halász 2008) requires closer comparisons within this species complex.

IV. Identification key for European species of *Alnus*-associated *Alpova* and *Melanogaster*

1a. Peridium (subpellis) pseudoparenchymatous. Gleba staining when cut (usually reddening). Spores ochre-yellow to pale grey-yellow sub micr., with narrow base < 1.5 µm wide, usually without remnants of sterigmata. Buffer cells always present aroung lacunae (genus *Alpova*) 2

1b. Peridium (subpellis) prosenchymatous. Gleba not staining when cut. Spores greyish-ochre sub micr., with large base > 1.5 µm wide, with conspicuous remnants of sterigmata. Buffer cells not present at maturity (genus *Melanogaster* sect. *Rivulares*) 4

2a. Surface glabrous, cream- to straw-yellow when young, smooth. Gleba reddening quickly (10-30 sec.) when cut. Dermatocystidia present (emergent and +/– thick-walled articles from peridiopellis). Spore width = 2.2-2.8 (3.2) µm, average volume 16-22 µm^3. Always under *Alnus alnobetula*, Alps, Carpathians, and Corsica, usually in organic soil. if without dermatocystidia, see also large-spored collections of *A. corsicus*) **1. *Alpova alpestris***

2b. Surface silky to slightly tomentous when young. Gleba reddening slowly (1-2 min.) or unconspicuously when cut. Dermatocystidia absent, but a tomentum early adpressed and collapsed with thin-walled terminal hyphae. Spore width = 1.8-2.5 µm, average volume = 10-14 µm^3. Under various *Alnus* in Corsica, *A. alnobetula* in the Alps, usually in mineral soil (sands or schists) 3

3a. In Corsica under *Alnus cordata*, *A. glutinosa* and *A. alnobetula* subsp. *suaveolens* (Corsica). Gleba deliquescent, slowly reddening **2. *Alpova corsicus***

3b. In the Alps with *Alnus alnobetula* subsp. *alnobetula*. Gleba not deliquescent, not distinctly reddening (only 1 basidiome observed !)
3. *Alpova* sp. (cf *A. cinnamomeus*)

4a. Basidiomata less than 1.5 cm diam., globose or flattened without distinct base and without pseudocolumella. Basidia predominantly 8-spored, wall not thickened. Spores 5.3-6.8 × 2.4-3.3 µm. With *Alnus incana* (and *A. glutinosa* ?) in alluvial substrates, Alps, Scandinavia **4. *Melanogaster luteus***

4b. Basidiomata more than 1.5 cm diam., with distinct base and short pseudocolumella usually distinct. Basidia often 10-spored, wall thickened at apex. Spores 5.1-6.2 × 2.7-3.1 µm. With *Alnus cordata* and *A. glutinosa*, Corsica
5. *Melanogaster rivularis*

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